VARIATIONS IN THE CORTICAL AND MEDULLARY BLOOD FLOW THROUGH THE DOG KIDNEY MEASURED WITH HEATED THERMOCOUPLES.

By

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Av

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This thesis is a summary of the following papers, which in the text below, e referred to by the Roman numerals.

GRÄNGSJÖ, G., SANDBLOM, J., ULFENDAHL, H.R. and WOLGAST, M., Theory of the heated thermocouple principle. *Acta physiol. scand.* 1966. *66.* 366–373.

. GRÄNGSJÖ, G., Regional blood flow in the kidney measured with heated thermocouples.

II. GRÄNGSJÖ, G. and PERSSON, E., The influences of some vasoactive substances on regional blood flow in the dog kidney. Acta anaesth. Scandinav. 1968. In press.

V. GRÄNGSJÖ, G., Autoregulation of regional blood flow in the dog kidney.

INTRODUCTION

One of the most important functions of the kidney is to regulate the water and salt balance, which it achieves by controlling the degree of concentration of the urine. It is generally accepted in the field of renal physiology that the final composition of the urine is determined during its passage through the collecting ducts towards the papilla. By analysis of micropunctates from Henle's loop it has been found that the osmolality of the urine increases in the direction of the papilla. According to a hypothesis originally proposed by Kuhn and Ryffel (1942) and later developed by Kuhn in collaboration with Wirz and Hargitay (1951), this successive increase in the osmolality is the result of a counter-current multiplier system in the medulla of the kidney. The driving force in this system is, according to Berliner et al.(1958) and Berliner and Bennet (1964), an active transport of sodium from the ascending limb of Henle's loop. During its transit through the medullary collecting ducts the urine is concentrated successively by means of a passive outward transport of water to the hypertonic interstitium. Further research will be required, however, perhaps especially with micropuncture techniques, before the ultimate solution of this problem is reached.

In the counter-current hypothesis the medullary blood flow is of critical importance. Thus a high medullary blood flow should "wash out" the hypertonicity created in the medulla.

In studies of the blood flow of the kidney, two functionally different concepts must be distinguished because of the counter current arrangement:

1) the absolute flow of blood cells and of plasma through a tissue region, and
2) the nutritive effect of the blood flow, i.e. the capacity of the flowing blood to transport substances to and away from a region.

Determinations of the absolute blood flow in the kidney were first made by Kramer and coworkers. By the analysis of dye dilution curves, intrarenally monitored, they calculated the blood flow in the cortex to be about 4.5 ml/min · g tissue, in the outer medulla about 1.1 ml/min · g tissue, and

in the inner zone of the medulla about 0.3 ml/min · g tissue (Kramer et al. 1960, Deetjen et al. 1964). Lilienfield et al. (1960) essentially confirmed the findings of the Kramer team of a low medullary blood flow, though their values for the medulla, calculated from the inflow rate of radioactively labelled albumin, were somewhat higher than those of Kramer et al. By internal monitoring of dilution curves from P^{32} labelled red cells with small betasensitive semiconductors, Wolgast (1968) found blood flows of essentially the same order of magnitude with respect to the cortex and outer zone of the medulla. For the inner zone of the medulla Wolgast calculated the flow to be 1.2 ml/min · g tissue, a value 3–4 times higher than those reported by Kramer and Lilienfield.

Nutritive blood flows in the kidney were measured by Thorburn et al. (1963) and Aukland (1964) by application of the inert gas technique of Kety and Schmidt (1945). With respect to the cortex and outer medullary zone of the kidney their values agree essentially with those obtained by measurement of absolute flows. In the inner zone, however, Aukland, with hydrogen electrodes, found flow values several times lower than those obtained by methods intended to give absolute flow values. This depends with certainty on the high diffusibility of the hydrogen gas, which is thus trapped in the counter-current system of the medulla. Despite this, however, Thorburn et al., using Kr^{85}, obtained flow values for the inner zone of the same magnitude as those reported by Kramer et al. and Lilienfield et al.

The thermocouple method used in the studies described in Papers I–IV can be compared with the inert gas method, as heat has a rather high diffusibility. However, this method offers some advantages over other methods, and used with criticism it can give valuable information on the regional blood flow in the kidney.

The purpose of this investigation was to study the *regional renal blood flow,* its changes at *different states of function* and at *different perfusion pressures,* and also the *influence of vasoactive substances.*

METHODOLOGY

The heated thermocouple

For measurement of the regional blood flow, heated thermocouples were used. The principles are well known and will therefore only be summarized briefly here.

One of the junctions in a thermoelement (in our case consisting copper-constantan) was heated with alternating current. The other junction lay at such a distance from the heated junction that it was not influenced by the heating. According to Grayson (1952) this distance should be 3–4 mm, when the thermocouples are applied to biological tissue. The heated junction thus constitutes the measuring point, and the other junction the reference point. The difference in temperature between these two junctions gives rise to a thermoelectric current, which can be recorded by a sensitive galvanometer.

When the thermocouple is placed in tissue the reference point thus assumes the temperature of the tissue, while the measuring point assumes a temperature dependent upon the amount of heat which is carried away both by conduction into the tissue and by convection through the blood flow. Changes in the blood flow in the vicinity of the measuring point can therefore be recorded as changes of the thermoelectric current. The region within which flow changes can affect the temperature of the measuring point constitutes, according to Grayson, a sphere with a radius of 3 mm.

In the animal experiments the heating current was chosen so that the tissue temperature was raised by a maximum of 2.5°C when no blood flowed past the measuring point. The copper-constantan junction gives 40 μV per °C, and with the galvanometers used this resulted in a 9 cm response. Thus very small changes in temperature could be detected. No amplifiers were necessary, and thus electronic drift was avoided.

Theoretical deduction (Paper I)

The heat transport from the heated measuring point consists of conduction and convection. Immediately adjacent to the thermocouple there is a non-perfused zone, caused by damage to the tissue on introduction of the thermocouple. There is also a certain distance between the thermocouple and the nearest blood vessels, which contributes to the size of the

non-perfused zone, through which heat transport will take place only by conduction. In the perfused medium the heat transport occurs by convection. Assuming that the surrounding medium had a homogeneous structure and that the heat flow from the probe was isotropic in all directions, the relationship was derived between the perfusing flow and the thermoelectric current (thermocouple response). This relationship was shown to be hyperbolic (eq. 9, I) when the thermocouple response was plotted against perfusing flow.

The hyperbolic relationship was demonstrated in model experiments. At high flow rates the temperature of the measuring point was equal to that of the reference point, indicating a negligible magnitude of the non-perfused zone.

THE RENAL STUDIES

According to findings presented by Fourman and Moffat (1964) the vascular pattern of the kidney seemed to suit the thermocouple method. Except for the cortico-medullary border there are no large vessels, but there is a well developed capillary system.

The experiments were performed on dogs. The responses from heated thermocouples in the cortex plotted against the blood flow in the renal artery fitted a hyperbola very well (I and II). This was considered to mean, in concordance with other authors, that the cortex takes the main part of the total blood flow to the kidney. Even at infinite flow there was a temperature difference between the measuring point and the reference point, indicating a non-perfused zone of no negligible magnitude. The responses from thermocouples located in the medulla, plotted in the same way, resulted in curves which appeared to reach a maximum rapidly. On infusion of papaverine, however, the response increased further with increasing flows in the renal artery. Thus the medullary flow had been restricted by a vascular tone which was abolished with papaverine. This was given the interpretation that the medulla, like the cortex has autoregulation of the blood flow. As expected, the calibration curve for a thermocuple in the cortex did not change on injection of papaverine. The calibration curves from thermocouples situated in the medulla were lower than those in the cortex. This was interpreted as an expression of a lower nutritive blood flow, under the assumption that the

haemorrhagic zone which was created on introduction of the thermocouple was of equal size in the cortex and medulla (II). Especially in the inner zone, it must be considered that the urinary flow and the counter-current arrangement may influence the thermocouple response.

Even if the response cannot be expressed in absolute values, the thermocouple method can give valuable information about the nutritive blood flow in different regions of the kidney.

Osmotic diuresis

During osmotic diuresis, induced by urea or mannitol, an unchanged or slightly reduced nutritive flow in the cortex was usually recorded (II). In the outer and inner zones of the medulla small increases in nutritive flows were found. These findings agree qualitatively with those of Thurau et al. (1960) and Wolgast (1968). Lilienfield et al. (1963), on the other hand, found decreased medullary blood flow on osmotic diuresis. The discrepancies may be explained merely by differences in their techniques, since all changes recorded are small.

Water diuresis

Water diuresis was rather difficult to obtain because of the anaesthesia and the operation trauma, but was studied on 3 dogs. Unsystematic changes were recorded in the cortex, and in the outer zone of the medulla the nutrient flow was unchanged or only slightly increased. Because of the very high diuresis, no definite conclusion could be drawn about the blood flow in the inner zone. No major changes were recorded either in the cortex or in the medulla. This is in opposition to Thurau et al. (1960), who calculated an increase of 70% in the blood flow, from the shortening of the mean transit time in their dye dilution curves. Changes of this magnitude should have been recorded with heated thermocouples. The discrepancies cannot be explained but need further investigation.

Vasoactive substances

Paper III describes the use of heated thermocouples to record the effects of vasoactive substances on the regional renal blood flow in normovolemic and hypovolemic dogs.

1. Pressor substances

Noradrenaline in continuous intravenous injections of less than 0.6 μg/min · kg body weight increased the blood flow in the renal medulla, and the diuresis, but decreased the cortical blood flow. These findings agree essentially with those of Langston et al. (1962).

All the other pressor substances tested, viz. adrenaline, methoxamine (Vasoxine^R), metaraminol (Aramin^R), normetaoxedrin (Effontil^R), Arg.^{8}-vasopressin (ADH) and angiotensin II (Hypertensin - N^R), produced decreased blood flow in the renal cortex and medulla. With the exception of ADH, these substances have not been tested by other authors in respect to the regional blood flow but only to the total blood flow to the kidney. In most cases a decreased total blood flow has been found, indicating that the renal vascular resistance was increased to a greater extent than the perfusion pressure. The effects of ADH on the regional renal blood flow, reported by Kramer et al. (in Thurau 1964) and Aukland (1967) agree with those described in Paper III; blood concentrations giving only an antidiuretic effect did not influence the blood flow. At higher concentrations the blood flow decreased in the cortex and medulla.

2. Vasodilative substances

Papaverine and bradykinin decreased the renal vascular resistance. Papaverine resulted in general vasodilation, and the net effect was a decreased renal blood flow. Bradykinin, however, with its short action time, gave increased renal blood flow in the cortex and the medulla without influencing the systemic blood pressure, when injected in the renal artery. It was also shown how bradykinin was able to eliminate extreme constriction of the renal vessels.

Carbon dioxide

Hypercapnia from breathing carbon dioxide mixtures or from apnoeic diffusion oxygenation (Holmdahl 1956), resulted in a reduction of the nutritive flow in the renal cortex and medulla and a decrease of the urinary flow. On apnoeic diffusion oxygenation the *systemic blood pressure was raised* probably because of the stimulatory effect of carbon dioxide on the

vasomotor centre. On breathing of mixtures of 5–20% carbon dioxide in oxygen, however, a *decreased arterial blood pressure* was recorded. It seems possible that the explanation lies in different depths of anaesthesia.

Haemorrhage and retransfusion

Bleeding resulted in a marked reduction of the nutritive flow, both in the cortex and the medulla, and the diuresis stopped. Retransfusion or infusion of plasma expander resulted in increased nutritive flow in all regions of the kidney and the existing anuria was abolished.

Autoregulation of renal blood flow

The autoregulation of the renal blood flow was studied (IV). In dogs anaesthetized with pentobarbital or chloralose the blood flow in the renal artery was fairly constant over the perfusion pressure range 90–220 mm Hg. With papaverine and also with bradykinin the autoregulation could be eliminated and the blood flow in the renal artery increased proportional to the increase in perfusion pressure.

The relationships between the perfusion pressure and the responses from heated thermocouples in the outer or inner zones of the medulla constituted S-shaped curves, with only small increases in response over the perfusion pressure range 80–250 mm Hg. This was considered to indicate the presence of autoregulation in the renal medulla. This opinion is supported by the findings of Aukland (1967) and Wolgast (1968).

GENERAL SUMMARY

(1) A theoretical deduction of the relationship between the fluid flow and the response of the heated thermocouple was shown to be a hyperbola. Experimental values from model and animal experiments fitted the theoretical curve quite well.

(2) Heated thermocouples were used in the dog kidney to study the regional blood flow and its changes at different states of function. On osmotic diuresis, the nutritive flow in the cortex decreased, but that in the medulla increased. On water diuresis only small and irregular changes were noted in all regions. Antidiuretic hormone in low concentrations produced no change

in the blood pressure or nutritive flow, but greatly increased the osmolality of the urine and decreased the urinary flow. At higher concentrations the nutritive flow decreased in all regions, and the blood pressure increased.
(3) The effects of some vasoactive substances on the regional renal blood flow were studied by means of heated thermocouples in normovolemic and hypovolemic dogs. Noradrenaline in continuous intravenous injections of less than 0.6 μg/min · kg body weight increased the blood flow in the renal medulla and the diuresis increased. All the other pressor substances tested, produced decreased medullary blood flow and decreased urinary flow. Small doses of bradykinin in the renal artery increased the blood flow in all regions of the kidney, without a decrease in the blood pressure. Hypercapnia resulted in a marked decrease of the blood flow in all regions and stopped the diuresis. On bleeding the dogs, there was a marked decrease in the blood flow to all regions and the diuresis stopped. Retransfusion or infusion of plasma expander increased the blood flow in all regions of the kidney and the diuresis started again.
(4) Autoregulation of the total blood flow was found in dogs anaesthetizied with pentobarbital or chloralose, but could be eliminated by papaverine or bradykinin. The relationships between the perfusion pressure and the responses from thermocouples situated in the outer or inner zone of the medulla were found to be S-shaped curves, indicating autoregulation of the blood flow also in the medulla. Sometimes, at perfusion pressures of 40–60 mm Hg, an increased thermocouple response was obtained, which might be interpreted as a redistribution of the renal blood flow, with increased medullary blood flow on hypotension.

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EXCERPTUM

Acta physiol. scand. 1966. 66. 366—373

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Theory of the Heated Thermocouple Principle

By

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Abstract

Grängsjö, G., J. Sandblom, H. R. Ulfendahl and M. Wolgast. *Theory of the heated thermocouple principle.* Acta physiol. scand. 1966. *66*. 366—373. — The heated thermocouple principle is widely used for blood flow measurements in tissues. A theoretical deduction of the relationship between the fluid flow and the response of the heated thermocouple is shown to be a hyperbola. Experimental values from model and animal experiments fit the theoretical curve quite well. It seems quite impossible to precalibrate the thermocouples due to the fact that they are surrounded by a nonperfused zone of injured tissue or noncirculating blood.

Gibbs (1933) introduced the heated thermocouple method for the measurement of regional blood flow and the method has since been used by many authors in different biological tissues, with more or less success (Linzell 1953, Grayson 1952). During the last few years Hensel (Hensel and Ruef 1954, Hensel and Bender 1956) has been one of the most eager advocates of the method.

The difficulty of calibration is the greatest disadvantage of the method. Most authors have studied the relationship between the "heat conductance" and the blood flow and have found the relationship to be linear (Dosekun, Grayson and Mendel 1960, Hensel and Ruef 1954, Hensel and Bender 1956, Graf and Rosell 1958). Other authors have found the "conductance" not to be proportional to the flow (Linzell 1953, Graf, Golenhof and Hensel 1957, Graf and Stein 1958). In his pioneer model experiments Bill (1962) discussed the nonlinear relationship between the "heat conductance" and the blood flow. He pointed out the impossibility of precalibrating the thermocouples in artificial media, as did Graf and Stein (1958) also.

In the present investigation the shape of the calibration curve of the heated thermocouples is deduced theoretically and the result is compared with that of model experiments. A simplified form of the theoretically deduced equation is given here, with reference to blood flow measurements in the renal cortex.

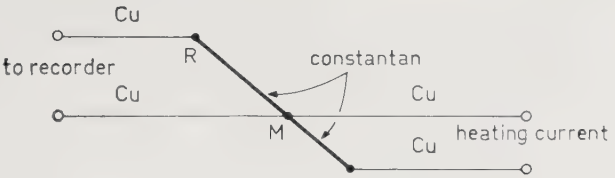

Fig. 1. The figure shows the principles of the thermocouple probe. Cu denotes copper wires. M is the heated point and R the reference point.

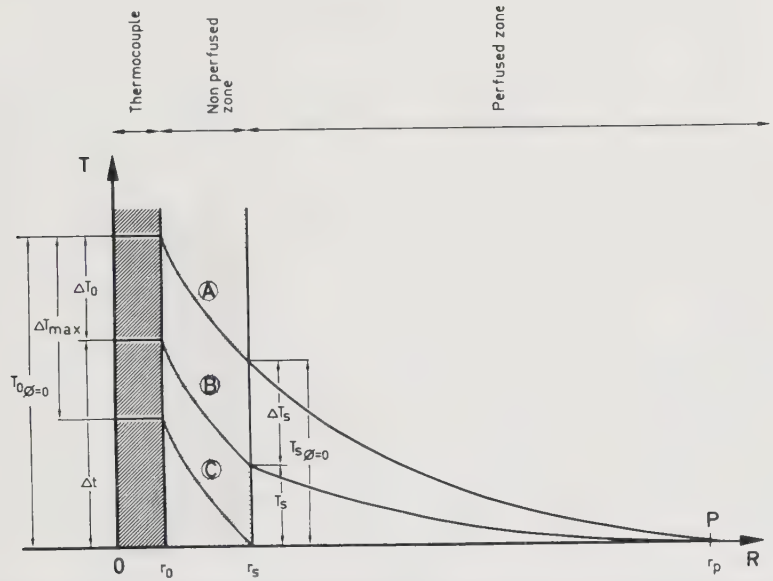

Fig. 2. The temperature profiles surrounding the thermocouple. T and R on the axis represent temperature and distance from the centre of the heated thermocouple, respectively. For explanation of the symbols see text. Curve A shows the profile at zero perfusion flow ($v = 0$) and curve C at infinite flow ($v = \infty$). Curve B represents an intermediate flow.

The performance of the heated thermocouple

The heated thermocouple consisted of a constantan wire about 7 mm long, 0.10 mm in diameter (Fig. 1). To each end an insulated copper wire of the same thickness was soldered. Two copper wires were soldered to a point on the constantan wire, dividing it into a 3-mm part and a 4-mm part. The short part was heated by an alternating current. The temperature difference between the two thermojunctions at the ends of the long part of the constantan wire was recorded. The probe used was similar to that described by Bill (1962).

Theory

Fig. 2 shows the temperature field surrounding the heated thermocouple in a tissue. The basal temperature of the tissue is set equal to zero. Immediately adjacent to the thermocouple appears a nonperfused zone, probably caused by damage to the tissue in introducing the thermocouple.

There is also a certain distance between the thermocouple and the nearest blood vessels, depending of the distribution of the vessels, and this contributes to the size of the nonperfused zone.

Heat transport will only occur through this nonperfused zone by conduction.

The following notation will be used:

r_o = radius of the thermocouple,
r_s = radius of the nonperfused zone,
r_p = distance from the centre of the thermocouple to the remote reference point, (point P in fig. 2),
T_o = Δt = temperature of the thermocouple,
T_s = temperature at the external surface of the nonperfused zone,
ΔT = $\Delta T_o = T_{o_{\Phi=0}} - T_o$, where $T_{o_{\Phi=0}}$ is the temperature of the thermocouple at zero flow,
ΔT_{max} = the value to which ΔT_o tends when the flow is increased infinitely and
T_{max} = $T_{o_{\Phi=0}}$ = the maximal temperature difference that can ever exist between the two thermojunctions with a certain heating current.

Curve A in Fig. 2 refers to the case when the perfusion velocity is made equal to zero and the transport of heat is pure heat conduction. Curve B shows the temperature profile at a moderate perfusion velocity and curve C the temperature profile at a very large perfusion velocity.

In the perfused medium the slope of the temperature profiles will in general be dependent on the flow, but in the nonperfused zone, where only conduction takes place, the profiles will become parallel, since the heat transfer is always the same if the heating current is kept constant Consequently the value of ΔT_o for an arbitrarily chosen flow will be equal at the surface of the nonperfused zone, $\Delta T_o = \Delta T_s$. ΔT_o and ΔT_s below are made equal to ΔT.

As seen in Fig. 2, ΔT_{max} (the decrease in temperature when the perfusion velocity flow tends to infinity) will be the same as $T_{s_{\Phi=0}}$ i.e. the surface temperature of the nonperfused zone at zero perfusion flow.

With certain assumptions about the surrounding medium, an expression relating the perfusion flow to the above-mentioned temperatures can be derived as follows.

In the first place, the medium is assumed to have a homogenous structure during perfusion. Secondly, the heat flow from the probe is assumed to be isotropic in all directions with the thermocouple at the centre of symmetry, and only radial components of heat flow need therefore be considered. Furthermore the reference point is regarded as being infinitely remote from the thermocouple.

The heat transport, $\dot{Q}$ (cal/sec), within the medium is composed of two quantities, namely, the conduction $\dot{Q}_K$ and the convection $\dot{Q}_V$, which together give the total heat flow

$$\dot{Q} = \dot{Q}_K + \dot{Q}_V. \tag{1}$$

The heat conduction $\dot{Q}_K$ is proportional to the gradient of temperature, where the constant K defines the coefficient of heat conduction and is expressed in cal/sec · cm.

$$\dot{Q}_K = -K\,\mathrm{grad}\,T, \tag{2}$$

where S is a surface surrounding the thermocouple.

The heat flow resulting from perfusion is obtained by multiplying the heat content of the medium with the radial component v of the perfusion velocity:

$$\dot{Q}_V = SvcT\varrho, \tag{3}$$

where ϱ denotes the density of the medium (g/cm³), c its heat capacity (cal/g · degree) and v its velocity (cm/sec).

Since both $\dot{Q}_K$ and $\dot{Q}_V$ are assumed to have the same direction, the total heat can be written:

$$\dot{Q} = -K\,\mathrm{grad}\,T + SvcT\varrho. \tag{4}$$

In a steady state the total heat flow across every surface which surrounds the probe is the same.

If R denotes the resistance of the heated part of the thermocouple and I the current passing through it, then the total heat flow, $\dot{Q}_o$, will be $0.24\ RI^2$ cal/sec.

Since the temperature profiles at high perfusion velocities extend a very small distance in comparison with the length of the probe, the geometry of the latter is best described as a long cylinder. The isotherms will then constitute cylinders with the radius r measured from the centre of the probe and with the surface S_r. S_r may be written as $S_s \cdot r/r_s$, where S_s is the external area of the nonperfused zone.

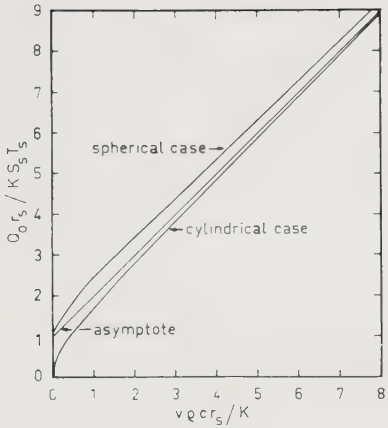

Fig. 3. A graphical representation of equation (7), including also the spherical theoretical case and the asymptote to these two equations. For explanation of symbols see text.

Equation (4) can now be written:

$$\frac{\dot{Q}_o r_s}{r} = -KS_s \frac{dT}{dr} + S_s v \varrho c T. \tag{5}$$

To solve this equation, both sides are divided by K and S_s and multiplied by the integrating factor $e^{-\frac{v\varrho cr}{K}}$, which gives

$$-\frac{\dot{Q}_o r_s}{KS_s r} \cdot e^{-\frac{v\varrho cr}{K}} = \frac{d}{dr} T \cdot e^{-\frac{v\varrho cr}{K}} \tag{6}$$

This expression can now be integrated between the following boundaries:

$T = 0$, $\quad T = T_s$;
$r = r_p = \infty$, $\quad r = r_s$.

Integrating equation (6) between these boundaries gives

$$\int_{r_s}^{\infty} -\frac{\dot{Q}_o r_s}{KS_s r} \cdot e^{-\frac{v\varrho cr}{K}} dr = \int_{r_s}^{\infty} \frac{d}{dr} T \cdot e^{-\frac{v\varrho cr}{K}} dr = -T_s \cdot e^{-\frac{v\varrho cr}{K}}. \tag{7}$$

The solution of this integral can only be obtained in terms of a series and must therefore be evaluated by numerical methods. If the integral is regarded as a function of v, it can be shown mathematically that the function has an asymptote for which equation (7) takes the form

$$\frac{\dot{Q}_o r_s}{KS_s} = T_s \left(1 + \frac{v\varrho cr_s}{K}\right). \tag{8}$$

By putting $v = 0$ on the right-hand side of equation (8), we get

$$\frac{\dot{Q}_o r_s}{KS_s} = T_{s\Phi=0}.$$

We also know that $T_{s\Phi=0} = \Delta T_{max}$ and $T_s = \Delta T_{max} - \Delta T$.

Substituting these relations and assuming v to be proportional to the total perfusion flow, Φ, equation (8) may now be written

$$\Delta T_{max} = (\Delta T_{max} - \Delta T)(1 + k\Phi), \tag{9}$$

where ΔT_{max}, according to equation (8) is $\dot{Q}_o r_s / KS_s$ and where T and Φ are measurable quantities and k a constant $\left(\propto \frac{\varrho c r_s}{K}\right)$. The relationship between ΔT and Φ, according to equation (9) is a hyperbola.

By plotting the quantity $\Delta T_{max}/(\Delta T_{max} - \Delta T)$ as a function of Φ, a straight line is obtained, according to equation (9) (Fig. 3).

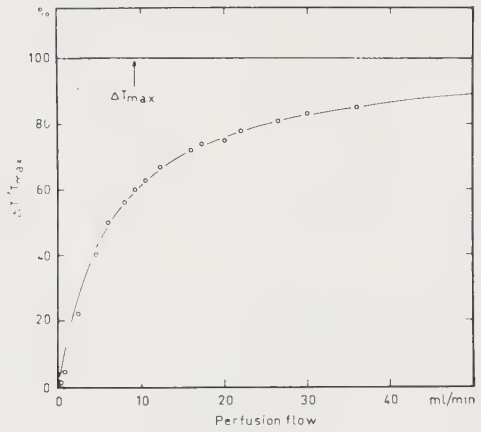

Fig. 4. The hyperbolic relation between perfusion flow and ΔT is shown. The open circles represent the experimental values and the curve the calculated hyperbola. ΔT is expressed as a percentage of T_{max}.

In the same figure the exact solution of equation (7) is represented, together with the solution of equation (4) for the case of spherical symmetry, i.e. when the isotherms are regarded as spheres. It can be shown that the spherical case has the same asymptote (expressed by equation (8)) as the cylindrical case.

For small perfusion flows the temperature profiles will extend further into the medium and the geometry is then expected to approach the spherical case.

Since in both cases the functions converge rapidly towards the asymptote, equation (8) may be considered approximately to describe the relationship between v and ΔT. If $\dot{Q}$ and v are taken to denote vector quantities, equation (4) is a very general equation and may, together with the steady-state condition div $\dot{Q} = 0$, be solved for any other set of conditions.

The assumptions made in deriving equation (9), however, are thought to be valid in most cases and experiments were made to test this theoretical model as below.

Experiments

Methods

Model experiments. A plastic tube, 13 mm in diameter, was filled with small glass beads varying between 80 and 100 μ in diameter. A Sigma pump forced water in a fairly steady flow through the system. The flow rate was controlled by a measuring cylinder.

The thermocouples — placed in the centre of the tube — were fed with the current from an AC power supply (Bill 1962). The potential difference between the reference and the heated thermojunction was recorded by mirror galvanometers on a photokymograph. All measurements were performed at room temperature.

Animal experiments. The heated thermocouples were used in studies on renal circulation in dogs (Grängsjö *et al.* 1966).

In anesthetized dogs the left renal artery was cannulated with a PVC tube and connected to the proximal end of one or both carotid arteries. The connection was kept at 37° C by a thermostatcontrolled water jacket. The blood flow was recorded by a recording rotameter (Shipley *et al.* 1951) and could be changed by clamping or by pumping with a Sigma pump.

The thermocouples were placed in the renal cortex by means of a needle soldered to two of the copper wires of the thermocouple. The wires were then soldered to a plug and connected to the heating device. The recording was performed as in the model experiments.

The blood flow through the cortex was regarded as being proportional to the flow in the renal artery (Kramer *et al.* 1960).

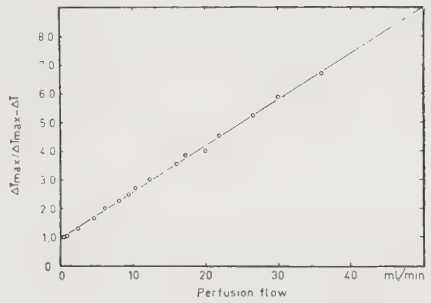

Fig. 5. The linear relationship between $\Delta T_{max}/(\Delta T_{max} - \Delta T)$ and perfusion flow. The open circles represent the experimental values.

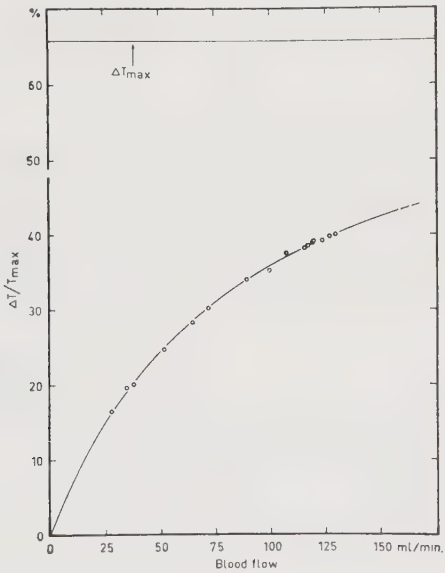

Fig. 6. The hyperbolic relation between total renal blood flow and ΔT. The open circles represent the experimental values and the curve the calculated hyperbola. ΔT is expressed as a percentage of T_{max}.

Results

Model experiments. In Fig. 4, ΔT from one experiment is plotted against the perfusion flow, Φ. The hyperbola, according to equation (9), was calculated by the method of least squares. In all model experiments T_{max} and ΔT_{max} coincided with each other, indicating a negligible nonperfused zone around the thermocouple. The linear relationship between $\Delta T_{max}/(\Delta T_{max} - \Delta T)$ and the perfusion flow is seen in Fig. 5.

Animal experiments. The results from the animal experiments were treated in the same way as those from model experiments. Fig. 6 shows the calibration curve from a renal experiment with the experimental points and the calculated curve. ΔT_{max} was always considerably lower than T_{max} and is drawn in the figure as the horizontal asymptote to the curve.

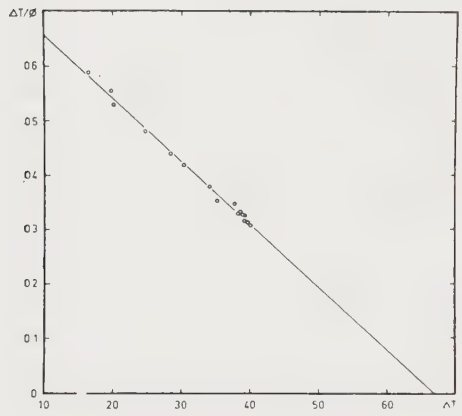

Fig. 7. The calibration line according to equation (10) with the thermocouple in the renal cortex. The values correspond to those in Fig. 6.

Equation (9) may be written

$$\Delta T/\Phi = k \cdot \Delta T_{max} - k \cdot \Delta T, \qquad (10)$$

representing a straight line, if $\Delta T/\Phi$ is plotted against ΔT.

When treated according to equation (10), the values in Fig. 6 give the result shown in Fig. 7.

The thermocouples have not been tested in other organs than the kidney.

Discussion

In order to deduce theoretically the relation between temperature changes, e.g. ΔT, and fluid flow, several assumptions were made. At least in the model experiments, the heat flow from the thermocouple was not actually the same in all directions but depended on the directed net flow given by the plastic tube. In spite of this, it seems to be valid to apply the experimental values to the derived formula (9). This seems also to be true of the renal cortex, where however, the blood flow is probably more uniform in all directions.

As stated in the beginning, ΔT is the temperature change of the heated thermojunction when the perfusion flow is increased above zero. This ΔT is not analogous to the temperature change Δt used in most work with heated thermocouples. Δt is the temperature difference between the reference junction and the heated junction (Fig. 2).

Most authors have not given the relationship between Δt and the flow. They have instead related ΔK to the flow, where ΔK means the difference of heat "conductivity" between the actual and zero flow. $1/\Delta t$ gives a measure of ΔK. The hyperbola representing this relation converges to a straight line when the nonperfused zone is very small. This may be a reason why some authors have found a linear relation between ΔK and the flow, while others have not. The deviation from the straight line is pronounced when there is a thick haemorrhagic zone or when the tissue is badly vascularized.

A linear relationship may also be explained by (1) the use of only the steep, relatively straight part of hyperbolic standardization curve, (2) the opening of new vessels or (3) the dilatation of vessels (according to Bill 1962).

It seems quite clear that the shape of the calibration curve must vary from one case to another, depending on the heat transport by conduction ($\dot{Q}_K$) or convection ($\dot{Q}_V$). *It is impossible to precalibrate, as generally the* $\dot{Q}_K$ *and* $\dot{Q}_V$ *can never be predetermined with sufficient accuracy. Thus the method requires calibration in situ.*

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REGIONAL BLOOD FLOW IN THE KIDNEY MEASURED WITH HEATED THERMOCOUPLES.

By

Göran Grängsjö

Uppsala 1968

ABSTRACT

Grängsjö, G., **Regional blood flow in the kidney measured with heated thermocouples.**

The regional blood flow in the kidney was studied in the dog with heated thermocouples, which, however, required calibration *in situ:* Calibrations were performed after elimination of the autoregulation of the kidney with papaverine. Results were obtained which indicated that the renal medulla also has autoregulation of the blood flow. On osmotic diuresis, induced by urea or mannitol, the nutritive flow in the cortex decreased, but that in the medulla increased. On water diuresis only small and irregular changes were noted in all regions. Antidiuretic hormone (Arg^8-vasopressin) in low concentrations produced no change in the blood pressure or nutritive flow, but greatly increased the osmolality of the urine and decreased the urinary flow. At higher concentrations the nutritive flow decreased in all regions, and the blood pressure increased.

INTRODUCTION

In the field of renal physiology there is general agreement that the final composition of the urine is determined during its transit through the collecting ducts towards the papilla. Cryoscopic studies of micropunctates from Henle's loop have shown that the osmolality of the urine increases in the direction of the papilla. According to the hypothesis of Wirz, Hargitay and Kuhn (1951), this is the result of a counter-current multiplier system, the driving force of which, according to Berliner et al. (1958), is an active transport of sodium from the ascending limb of Henle's loop, the urine being concentrated by means of a passive outward transport of water from the medullary collecting ducts into the hypertonic interstitium. This theory has found fairly general acceptance, but there has been some criticism. Marsh and Solomon (1965) obtained results in perfusion experiments, indicating that Henle's loop is not capable of active salt transport, and from a later study (Marsh 1966) it was concluded that the driving force for concentration of the urine was an active water transport in the medullary collecting ducts. Knowledge of the localization of the active process of urinary concentration will require further research, especially with microtechniques. It is probably, however, in view of the correlation that has been found between the length of Henle's loop and the ability for urinary concentration in different mammals, that this loop takes an active part in the concentration process. Regardless of the localization of the driving force underlying the ability of the kidney to build up an increasing osmolality in the direction of the papilla, Henle's loop and the vasa recta constitute a system whose task it is to preserve this hypertonicity. It is of critical importance that the blood flow in the vasa recta to this region, the inner medulla, should be so low that the concentration gradient which is built up from the cortico-medullary border to the papilla is not "washed away". Attention was drawn to the importance of the vasa recta system by Hargitay and Kuhn as early as in 1951, and several investigators have attempted, by different methods and with varying degrees of success, to determine the medullary blood flow.

Kramer and coworkers (Kramer et al. 1960, Thurau et al. 1960, Deetjen et al. 1964 and Meier et al. 1964), using an indicator dilution technique, made simultaneous determinations of the blood flow in the cortex and medulla of the kidney, and found large differences between these two regions. The blood flow in the cortex was found to be 4.5 ml/min · g tissue, while that in the outer zone of the medulla was 1.1 ml/min · g tissue and in the inner medullary zone it was as low as 0.3 ml/min · g tissue (in Thurau 1964). Further, these authors claimed to have shown that the medullary blood flow increased both in water diuresis and osmotic diuresis, and that an autoregulatory mechanism existed in the cortex but not in the medulla.

Lilienfield and Maganzini (1960), by studying the inflow rate of radioactively labelled albumin, calculated the blood flows of the different regions of the kidney, and confirmed the findings of the Kramer team that there was a relatively small blood flow in the medulla as compared with the cortex. In osmotic diuresis, however, in contrast to the results of Kramer et al., Lilienfield and Maganzini (1960) found a decrease in the blood flow of the medulla.

In a previous paper we reported determinations of the regional blood flow by means of semiconductor detectors (Grängsjö, Ulfendahl and Wolgast, 1966). A detailed description of the method, together with biological results in the form of blood flow values in the cortex and medulla of the kidney under different physiological conditions, has recently been published by Wolgast (1968).

In spite of numerous investigations on the regional blood flow in the kidney, many questions remain unanswered, and many factors are still obscure. This is largely due to the fact that for different reasons the various methods used have been unsatisfactory, which in turn is due to the complex flow conditions prevailing in the kidney.

In studies of the blood flow of the kidney, two functionally different concepts must be distinguished. Thus studies can be made of the *absolute flow of blood cells and/or plasma* through a tissue region, or of the *nutritive effect of the blood flow,* i.e. the capacity of the flowing blood to transport substances to and away from a region. Kramer et al., Lilienfield et al. and Wolgast have studied the *absolute flow,* while with thermocouples and inert gas methods (cf. Thorburn 1963, Aukland 1964) it is rather the *nutritive blood flow* that is measured.

The present paper reports results from flow measurements in the kidney made with the aid of heated thermocouples, as according to the principle of Gibbs (1933). Measurements of regional blood flows with heated thermocouples have been reported previously by many workers, mainly in skin and muscle, but the method has also been used in the kidney, for example by Scher (1951) and Perl et al. (1965), the latter authors using a method modified as described by Stow et al. (1959).

Some investigators, including Mowbray (1959), have attempted to precalibrate their thermocouples, but on the basis of a previous study (Grängsjö et al. 1966) this was not considered feasible (cf. Bill 1962). Thus each thermocouple has to be calibrated *in vivo*, which obviously entails great difficulties, unless reference methods are available. The thermocouple method offers advantages over other methods, however, by its simplicity and, above all, because it allows continuous recording of the flow, whereby rapid and transient changes can be detected. In all other methods reported in the literature the determinations can only be made at stationary flow rates.

The aim of this study was to investigate the potentialities of thermocouples for blood flow recordings in different parts of the kidney, and the possibilities of calibration of the response. Then the method was applied to studies of blood flow changes in osmotic diuresis, water diuresis and antidiuresis.

EXPERIMENTS

Methods

Anaesthesia. The experiments were performed on a series of 30 dogs, varying in weight between 6.5 and 32 kg. They were allowed free access to water until the start of the experiment. In the experiments at the beginning of the series the dogs were anaesthetized with pentobarbital sodium i.v. in a dose of 30 mg/kg body weight, but it was found impossible to produce water diuresis in these dogs, probably due to the fact that this anaesthetic agent, like all other barbiturate preparations (Sawyer 1961), causes excess secretion of ADH, and in later experiments a 1 per cent chloralose solution i.v., in a dose of 50 mg/kg body weight, was used usually after induction with thiopentone sodium (Pentothalsodium^{R}, ABBOT)★ i.v., in a dose of 20 mg/kg body weight. The anaesthesia was maintained with a 1 per cent solution of chloralose, 30–40 ml per hour. All dogs were intubated tracheally.

Operation technique. The anaesthetic and other substances were administered through a polythene catheter inserted in a superficial vein in the leg. The mean systemic blood pressure was recorded by means of a catheter inserted in the femoral artery and connected to a pressure transducer. The left kidney was exposed through a flank incision, and the renal artery was dissected free, after which it was connected to the common carotid artery or the femoral artery by means of a tube system thermostatically controlled at 37°C. The blood flow to the kidney was recorded with a rotameter (Shipley et al. 1951) and could be deminished by clamping or changed by pumping with a Sigma pump, after which a "wind-kessel" was connected. The dogs were heparinized with a dose of 15–20 mg/kg body weight. Injections could be made directly into the renal artery through the tube system. The pressure with which the kidney was perfused was recorded via a pressure transducer. The urine was collected by means of catheters introduced into the ureters, and the urine flow recorded by a drop counter which was eletronically reset after each 30th drop. 30 drops had a volume of 1 ml.

★ Pentothal-sodium was kindly supplied by Abbot AB, Stockholm, Sweden.

Heated thermocouples

The thermocouples used here were of the same construction as those described in a previous paper (Grängsjö et al. 1966), and consisted of a constantan wire about 7 mm long and 0.10 mm in diameter. To each end an insulated copper wire of the same thickness was soldered. Two copper wires were soldered to a point on the constantan wire, dividing it into a 3 mm and a 4 mm part. The shorter part was heated by a stabilized alternating current (50 c/s), which was chosen so that the tissue temperature was raised maximally about 2.5°C, when no blood flowed past the thermocouple. At the usually existing blood flows in the renal parenchyma the temperature rise amounted to 0.5–1°C. The wires leading to the heated part were soldered end-to-end to a needle 0.25 mm in diameter, which was pushed thorugh the kidney and the thermocouple could then be drawn to the desired location in the renal parenchyma. In each experiment five thermocouples were usually introduced, at different levels in the kidney, and their exact positions were determined by careful dissection after fixation for about 24 hours in 6 per cent formalin solution, when the different zones of the kidney were easily distinguishable. At this dissection a number of thermocouples were excluded from the experiments, particularly those whose measuring points lay close to large vessels and thereby had a low sensitivity (cf. Bill 1962). This occured especially in the corticomedullary border with its arcuate vessels. Other thermocouples were excluded and replaced during the actual course of the experiment, namely those which were found to be unstable and which reacted violently on injection of cold physiological saline into the renal pelvis. This indicated that the measuring point lay close to or in the renal pelvis, a possibility which was verified at the dissection. The thermo-electric current caused by the temperature difference between reference point and measuring point was recorded with a mirrorgalvanometer on a UV recorder. Because of the high sensitivity of the galvanometers amplifiers were not required, and the responses from the thermocouples were thus not subjected to any electrical drift. This was a necessary condition, since many of the experiments lasted for 12–14 hours. With the UV recorder the systemic blood pressure, perfusion pressure, the blood flow in the renal artery and the urinary flow were also recorded.

Osmotic diuresis was induced by infusion of 10% urea solution or 15% mannitol solution (Mannidex^{R}, AB Pharmacia, Sweden).

Water diuresis was induced by infusion of 0.45% NaCl solution, which gives no obvious haemolysis of dog red cells (Thurau, Deetjen and Kramer 1960). In the experiments with water diuresis great care was taken to arrest haemorrhage, since this is perhaps the strongest stimulant to the release of antidiuretic hormon (Sawyer 1961). In spite of the oral administration of large quantities of water and 0.45% saline solution i.v., water diuresis could not be induced in dogs anesthetized with pentobarbital sodium. On the other hand, in three dogs in which anaesthesia was induced with thiopentone sodium and maintained with chloralose, and in which bleeding was carefully arrested at the operation, hypotonic urine was obtained.

Antidiuresis was induced by infusion of Arg^8-vasopressin★, the specific antidiuretic hormone for dogs (Sawyer 1961).

Urine osmolality was determined by measurement of the freezing point reduction (Osmometer 31 LS Advanced Instruments, USA).

RESULTS

Calibrations

In a previous study (Grängsjö et al. 1966) it was shown, in model experiments and for the renal cortex, that the calibration curve for a thermocouple is hyperbolic.

$$\Delta T = \frac{\Delta T_{max} \cdot \phi}{\frac{1}{k} + \phi} \quad , \qquad (1)$$

where ΔT is the change in temperature difference between the measuring and reference points when the flow increases from zero to some particular flow rate (Fig. 1). ΔT increases with increase of flow to a limiting value ΔT_{max} as the asymptote of the hyperbolic calibration curve is reached at infinite flow rates. T_{max} is the change in temperature difference between measuring point and reference point when the flow is zero and the heating current is switched on and off, respectively. ΔT is expressed in per cent of T_{max}; ϕ is the cooling blood flow past the heated soldered junction (measuring point), and k a constant which comprises the density of the tissue, the heat capacity, the radius of the non-perfused zone and the coefficient of heat conduction.

★ Arg^8-vasopressin was kindly supplied by AB Ferring. Malmö, Sweden.

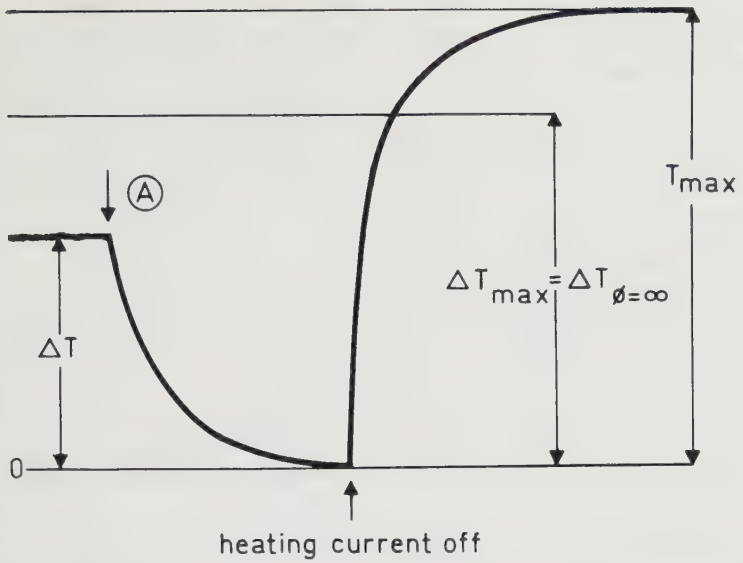


Figure 1.

The figure shows how the thermocouple response, ΔT, decreases to 0 when the dog dies (at arrow A). The temperature difference between the measurement and reference points of the thermocouple is then greatest. When the heating current is switched off, the measurement and reference points assume the same temperature, whereby the value T_{max} is reached asymptotically. ΔT_{max} is determined graphically (see text).

When used in tissue, the magnitudes of ΔT_{max} and k will differ depending on the location of the thermocouples in relation to blood vessels, and perhaps especially on the pattern of the capillary network. Also of great importance is the size of the non-perfused zone in the immediate vicinity of the thermocouples. *It is thus not feasible to calibrate the thermocouples other than at the site of measurement.*

In the calibration, ΔT was plotted against the total flow to the kidney, measured in the renal artery. The blood flows in the different regions of the kidney were assumed to be proportional to the flow in the renal artery when all myogenic mechanisms in the vascular system of the kidney had been eliminated. This was produced by continuous infusion of papaverine into the renal artery (Thurau and Kramer 1959), 0.8–1.1 mg/min · kg.

It was found that the calibration curve for a thermocouple in the cortex

was the same whether the myogenic mechanism was eliminated or not (Fig. 2). When ΔT from the thermocouple in the medulla was plotted against the total flow in the renal artery, a curve was obtained which appeared to rapidly reach a maximum (Fig. 2), but on infusion of papaverine the respon from the medullary thermocouple increased further with increasing flows in the renal artery.

Equation (1) can be written

$$\frac{\Delta T}{\phi} = k \cdot \Delta T_{max} - k\Delta T \quad (2)$$

which corresponds to a straight line if $\frac{\Delta T}{\phi}$ is plotted on the ordinate and ΔT on the abscissa. When $\frac{\Delta T}{\phi} = 0$, $\Delta T = \Delta T_{max}$ and ΔT_{max} can thus be mined graphically as the x-intercept. In calibrations where the regu-

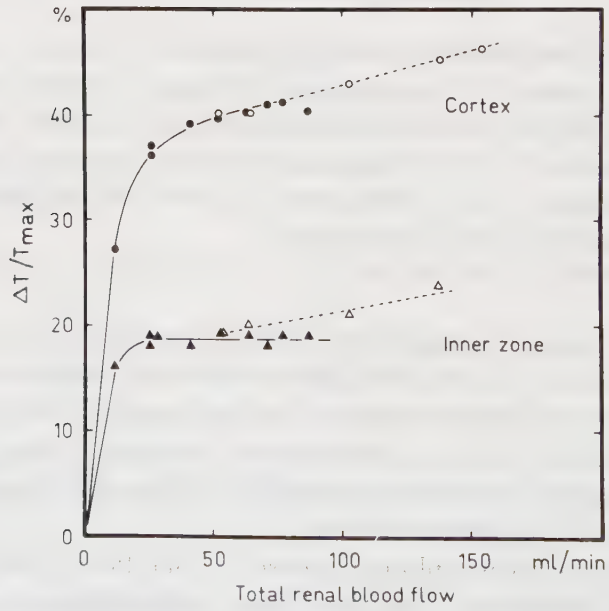


Figure 2.

Calibration curves from the cortex and inner zone of the medulla. The fille circles and triangles represent calibration without papaverine, while the unfilled circles and triangles refer to continuous infusion of papaverine into th renal artery.

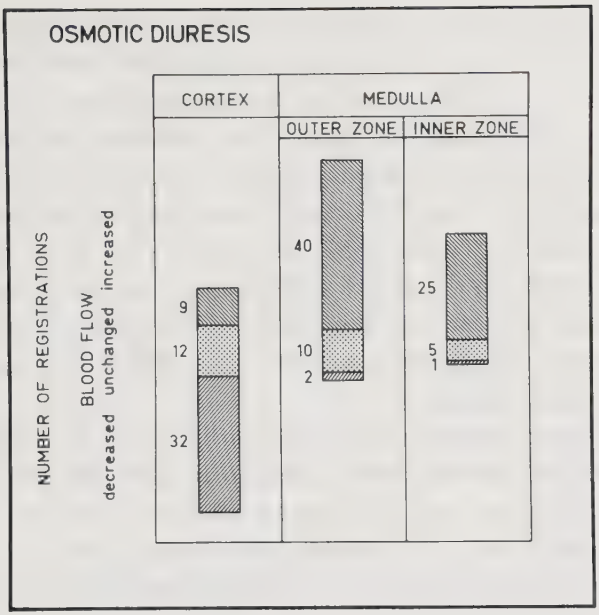


Figure 3.

Summary of blood flow changes recorded during osmotic diuresis induced by urea or mannitol.

latory mechanism was eliminated by papaverine, ΔT_{max} was determined graphically, and for thermocouples in the cortex usually lay between 50% and 60%, for those in the outer medullary zone between 35% and 50%, and for those in the inner medullary zone between 20% and 30% of T_{max}.

It was sometimes found that the thermocouple responses decreased during the course of the experiment, probably because the non-perfused zone, the haemorrhagic zone, increased. For this reason, comparison between responses from the same thermocouple could not be made with certainty if too long a time had elapsed between observations.

Osmotic diuresis

Osmotic diuresis was induced in a total of 18 dogs, in 15 with urea and in 4 with mannitol. The amount of infused urea varied between 1.5 and 11 g, and the increase in diuresis was proportional to the amount injected. On

infusion of the urea solution a slight increase in blood pressure of 5–10 mm Hg usually occurred. The amount of mannitol infused varied between 1.5 g and 10.5 g. It had no effect on the blood pressure.

The results are illustrated in Fig. 3. The pattern is clear and may be interpreted as meaning that osmotic diuresis gives a reduction of the blood flow in the cortex (32 out of 53 cases) but an increase in the outer and inner zones of the medulla (40 out of 52 cases, and 25 out of 31, respectively). Nine thermocouples in the cortex recorded an increased flow. In these 9 cases the osmotic diuresis was induced by urea, which caused an increase in the blood pressure. In other respects the results from urea and mannitol diuresis were in agreement.

The increased thermocouple responses which were recorded in the medulla have been interpreted as being due to an increased blood flow, but in the case of the inner zone the increase may have been due partly to an increased urinary flow in the tubules and collecting ducts.

In two experiments in which the total flow to the kidney was measured

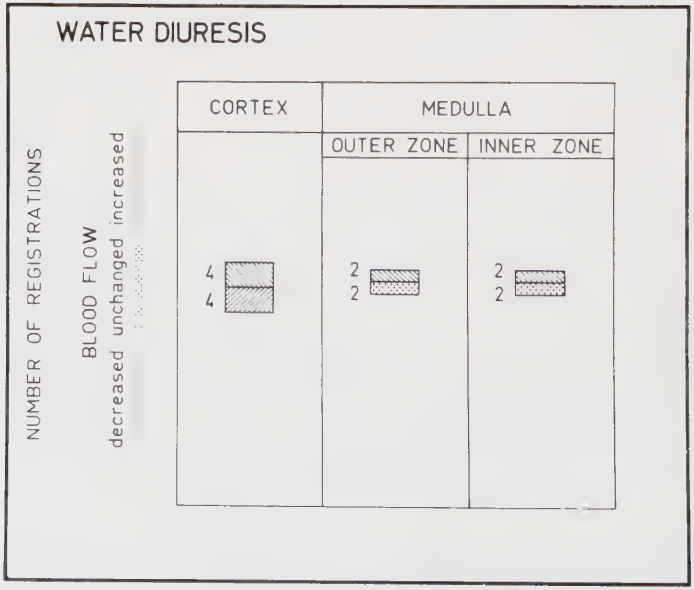


Figure 4.

Summary of blood flow changes recorded during water diuresis.

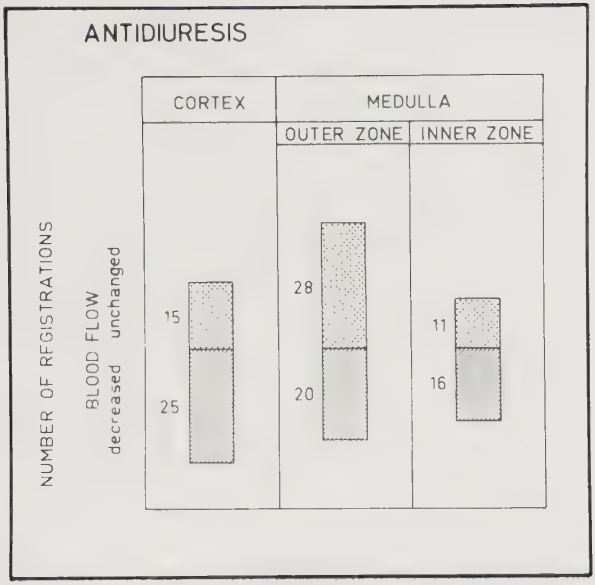


Figure 5.

Summary of blood flow changes recorded during antidiuresis induced by Arg^8-vasopressin.

in the renal artery with a rotameter, a negligible decrease in flow was found during osmotic diuresis.

Water diuresis

The effect of water diuresis was studied in 3 dogs. After the oral administration of 600–800 ml water and 500–900 ml 0.45% saline solution intravenously, a large increase in diuresis was obtained, and the urine osmolality decreased to 150–190 mosm/kg. Out of 8 thermocouples in the cortex, 4 gave a somewhat lower response than the initial value (Fig. 4.). Out of 4 thermocouples in the outer zone, 2 showed an unchanged response and 2 a negligibly higher response than previously. Two thermocouples in the inner zone showed no change, while 2 others increased their responses parallel with the increase in diuresis.

Antidiuresis

Arg^8-vasopressin was given to 6 dogs for induction of antidiuresis, in doses varying between 0.0017 and 0.16 IU/min · kg intravenously.

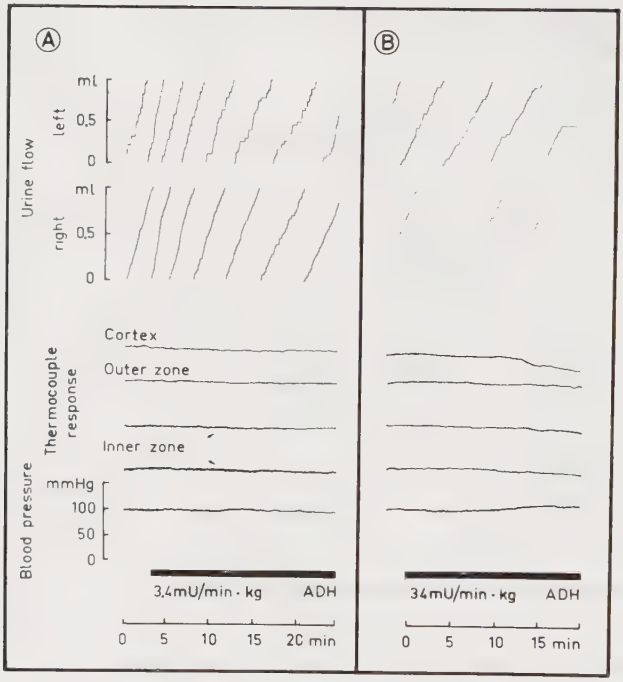


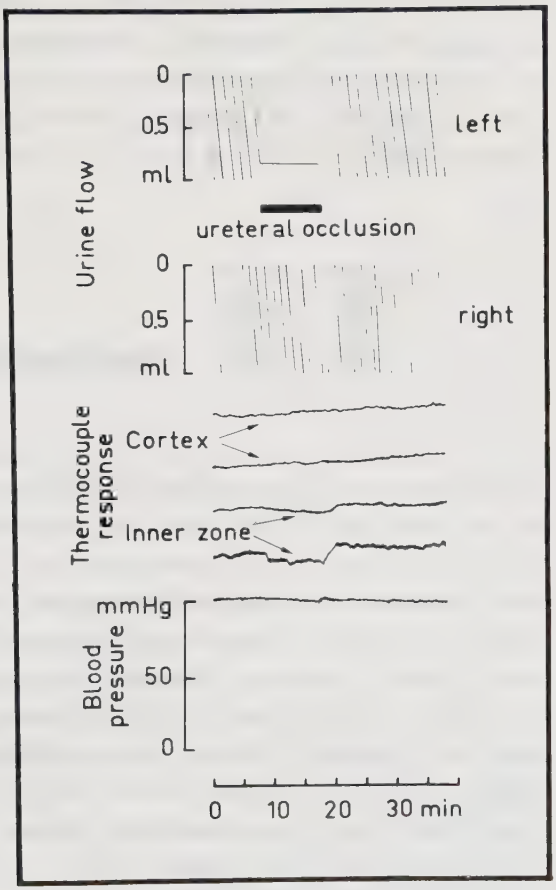
Figure 6.

The figure shows (A) that a low concentration of Arg^8-vasopressin on continuous infusion had no influence on the thermocouple response or on the systemic blood pressure, but decreased the urinary flow by about one half. At high concentrations (B) the blood pressure increased, and the thermocouple response and diuresis decreased.

A rapidly initiated decrease in diuresis and increase in the osmolality of the urine were observed. At the high doses, urine osmolality values of about 1000 mosm/kg were obtained. At the lower doses no effect on the blood pressure was noted, but at the higher doses the systemic blood pressure increased by 10–20 mm Hg.

The results are illustrated in Fig. 5. In the case of the cortex there was a decrease in flow in 25 out of 40 cases, and no change in the remaining 15. In the outer zone of the medulla, no changes in flow were observed in 28 out of a total of 48 recordings, while in the remaining 20 recordings the flow decreased. In the inner zone no change in flow was recorded in 11

ne figure shows changes in thermocouple response on ureteral occlusion
or 10 minutes. The nutritive flow in the cortex increases slightly. The
hermocouple responses in the inner zone are lower during the occlusion,
nd rise practically to the initial value when the occlusion is released.



igure 7.

out of 27 cases, while the flow decreased in 16 cases. For both the cortex and medulla it was found that low doses, which had no effect on the blood pressure but did increase the urine osmolality, resulted in no flow changes (Fig. 6). The higher doses, on the other hand, increased the blood pressure by 10–20 mm Hg and decreased the blood flow in the cortex and medulla.

Stop flow

In an attempt to ascertain to what extent the degree of diuresis, i.e. the urinary flow in the tubules and collecting ducts, influenced the response of the thermocouples, the ureters were occluded or a pressure of 100 mm Hg was applied in the renal pelvis. For thermocouples lying in the cortex or the outer zone of the medulla the responses were either unchanged, or the changes were small and irregular. On the other hand, the responses of thermocouples in the inner zone of the medulla always decreased (Fig. 7).

DISCUSSION

Measurement of the absolute blood flow in different regions of the kidney has been made by Kramer and coworkers (Kramer et al. 1960, Thurau et al. 1960, Deetjen et al. 1964, Meier et al. 1964) and also by Lilienfield and coworkers (Lilienfield and Maganzini, 1961, Lilienfield and Pomerantz, 1963). The results obtained are in essential agreement as regards flows under normal conditions. Flows of the same order of magnitude have also been obtained by Wolgast (1968) with respect to the cortex and the outer zone of the medulla. For the inner medullary zone, on the other hand, Wolgast calculated the mean flow to be 1.2 ml/min · g tissue, a value 3–4 times higher than those reported by Kramer and Lilienfield (0.38, and 0.2–0.6 ml/min · tissue, respectively).

Nutritive flows in the kidney were determined by Aukland (1964), and his values agree essentially with those of the above authors for absolute blood flows in the cortex and outer medullary zone. Because of the counter-current system in the inner zone of the medulla and the urinary flow in the collecting ducts, Aukland obtained flow values for this region which were several times lower than those obtained with methods which are intended to give absolute flow values. It may be asked, however, whether his values are not to some extent elucidative of the nutritive conditions in the inner zone. The same objections as regards the inner zone

can be raised against the method used by Thorburn et al. (1963), where Kr^{85}, like hydrogen gas, is trapped in the counter-current system and only to a minor extent reaches the inner zone and papilla. Despite this, the flow value obtained by Thorburn for the inner medullary zone of the kidney agree with those obtained by Kramer and Lilienfield.

The aim of the present investigation was to study the blood flow of the kidney with heated thermocouples and to elucidate further the nutritive flow conditions in different regions of the kidney, particularly in the medulla.

The method of using thermocouples has the advantage, as opposed to the methods mentioned above, that it allows continuous measurements, so that rapid and transient changes can be recorded. In a previous study (Grängsjö et al. 1966) it was shown that thermocouples are applicable in *tissues with a homogenous blood flow,* through which no large vessels run (cf. Bill 1962). It is considered that the different regions of the kidney, with the exception of the cortico-medullary region, well fulfil these criteria as regards the vascular pattern, which has been described previously by v. Möllendorff et al. (1930) and in more recent years by Fourman et al. (1964).

Calibrations

Since, according to previous findings, the thermocouples had to be calibrated *in situ*, a series of calibrations were performed, in which, in complete accordance with the findings in model experiments, it was found that the calibration curve for thermocouples in the cortex consisted of a hyperbola when the thermocouple response, ΔT, was plotted against the flow in the renal artery (Fig. 2). This calibration curve showed no noteworthy change when the myogenic mechanism in the vessels had been eliminated with papaverine, and this was given the interpretation that the cortical flow comprises the major part of the flow in the renal artery and is essential proportional to the total flow, in complete agreement with the findings of other investigators.

After elimination of the autoregulatory mechanism, calibration curves for thermocouples in the medulla were obtained, in which ΔT increased with increasing flows in the renal artery, which was considered to mean that the medulla, also has autoregulation. This contrasts to the views of Thurau

et al. (1959) but is in agreement with those of Aukland (1967) and Wolgast (1968). Further investigations of the autoregulation of the kidney will be reported in a later paper (Grängsjö 1968).

It was a consistent finding in the papaverine experiments that the calibration curves from the cortex lay at a higher level than those from the medulla.

For the cortical thermocouples ΔT_{max} usually lay between 50 and 60%, for the thermocouples in the outer medullary zone between 35 and 50%, and for those in the inner zone between 20 and 30% of T_{max}. A lower ΔT_{max} was interpreted as an expression of a lower nutritive blood flow, under the assumption that the haemorrhagic zone which was formed on introduction of the thermocouple into the kidney was of equal size in the cortex and medulla. At the dissections for establishment of the exact locations of the thermocouples, no differences were found, as a rule, in the sizes of these haemorrhagic zones. When such a difference did exist, then the haemorrhagic zone in the cortex was larger, which would give a too low value for ΔT_{max} for the cortex.

Because of the length of the thermocouple it cannot be used for recording from the deepest parts of the inner medulla or the papilla, since in these cases the measuring point is too close to the renal pelvis, and in addition, owing to the large number of collecting ducts the urinary flow can have an effect and give a false increase of ΔT. On ureteral occlusion, no or only a negligible effect of ΔT was found for thermocouples in the cortex and outer zone of the medulla. On the other hand ΔT decreased for thermocouples in the inner zone. This decrease was probably due to the cessation of urinary flow, but also to a reduced medullary blood flow (Kramer et al., cited Thurau 1964). The fact that the urinary flow can influence the thermocouple response can be said, however, to support further the concept of a low nutritive blood flow in the inner parts of the medulla. That the magnitude of ΔT for thermocouples in the inner zone does not depend exclusively on the urinary flow in the collecting ducts, was evident after the administration of small doses of adrenaline (Grängsjö and Persson 1968); the diuresis then decreased, but at the same time ΔT increased for thermocouples in the inner medullary zone, which was regarded as an expression of increased nutritive flow (cf. Aukland and Krog 1961, Ulfendahl 1962). In measurements with thermocouples in the medulla, especially in the inner

zone, it is certain that, as in the case of the inert gas methods, the countercurrent arrangement has some influence, and that heat produced in the measuring point will to a certain extent be retained in the region. If, however, it is assumed that the capacity of the blood flow to carry away heat is proportional to its nutritive capacity, the thermocouple method can give valuable information about the nutritive blood flow in different regions of the kidney, even if ΔT cannot be expressed in absolute values.

Osmotic diuresis

During osmotic diuresis, an unchanged or slightly reduced nutritive flow in the cortex was usually recorded. The nine cases in which an increase in flow was recorded in the cortex all referred to experiments in which the osmotic diuresis was induced by urea, which increased the systemic blood pressure. In six of these nine cases the autoregulation had been previously eliminated by an injection of papaverine, and the increase in blood pressure was therefore probably the cause of the increased flow. In the outer and inner zones of the medulla an increased nutritive flow was found. This is in agreement with Thurau et al. (1960), who with their method recorded a shortening of the mean transit time for indicator dilution curves from the medulla, which was interpreted as an expression of an increased blood flow. Our finding contrasted, on the other hand, with the results reported by Braun and Lilienfield (1963). In a study on the blood flow of the kidney Wolgast (1968) has presented indicator dilution curves for P^{32}-labelled red cells on internal recording with beta-sensitive semiconductors. In agreement with Thurau et al. (1960), he found a shortening of the mean transit time during osmotic diuresis with mannitol, but since at the same time he found that the blood volume decreased, he concluded that osmotic diuresis caused no major changes in the blood flow.

It was concluded from measurements with thermocouples that in osmotic diuresis the nutritive flow to the renal cortex is unchanged or very slightly reduced, while the nutritive flows to the outer and inner zones of the medulla are increased.

Water diuresis

In water diuresis with urine osmolalities of 150–190 mosm/kg, i.e. water diuresis of the same order of magnitude as in the study of Thurau et al.

(1960), no or only negligible changes in the thermocouple response, which was considered to mean that there were no or only negligible changes in the regional blood flow. The finding of Thurau et al. (1960) of a shortening of the mean transit time, in the case of the medulla, to approximately one half, indicates large changes in flow, and these authors calculated an increase in the blood flow of 70%. Such large increases should be detectable without difficulty with thermocouples lying in the outer zone. It is difficult to make a definite statement about the blood flow in the inner zone, since in these experiments the diuresis was very high, and the urinary flow would certainly have influenced the magnitude of ΔT. It is probable, however, that no large changes of the nutritive flow took place in the inner zone.

Antidiuresis

No marked decrease in the regional blood flow of the kidney was found on infusion of antidiuretic hormone in physiological concentrations. Concentrations were regarded as physiological when they gave decreased diuresis and hypertonic urine without affecting the systemic blood pressure. When, on the other hand, the concentration of vasopressin was increased reduced blood flows were recorded in all regions in spite of an increased blood pressure. The vasopressin appeared thus to have increased the resistance in the vessels of the kidney, with decreased renal blood flow as a result, an effect which is probably obtained from most pressor substances (Grängsjö and Persson 1968).

In a large number of unpublished experiments, Kramer and coworkers (cited Thurau 1964) found no vasoactive effects of antidiuretic hormone in physiological concentrations, while with high concentrations results were obtained which indicated a vasoconstriction of the vessels in both the cortex and the medulla. The same result was obtained by Aukland (1967), who found no obvious decrease of the hydrogen gas clearance for the outer zone of the medulla on infusion of antidiuretic hormone. Neither does Thorn (1968) consider it probable that antidiuretic hormone in physiolgical doses has any influence on the regional blood flow in the kidney.

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In press

THE INFLUENCES OF SOME VASO-ACTIVE SUBSTANCES ON REGIONAL BLOOD FLOW IN THE DOG KIDNEY.

by

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SUMMARY

Changes in the renal blood flow, with special reference to the regional blood flow, were studied in normovolemic and hypovolemic dogs. Heated thermocouples, *ad modum* Gibbs, were used. The effects of adrenaline, noradrenaline, methoxamine, metaraminol, normetaoxedrin, Arg^8-Vasopressin and angiotensin II were studied. Noradrenaline in continuous intravenous injections of less than 0.6 μg/kg · min. increased the blood flow in the renal medulla and the diuresis increased, but the cortical blood flow decreased. Other pressor substances produced decreased blood flow in the renal cortex and medulla. Injection of papaverine resulted in decreased arterial blood pressure through general vasodilatation and the renal blood flow decreased. Small doses of bradykinin in the renal artery increased the blood flow in all regions of the kidney, without a decrease in blood pressure. The effects of apnoeic diffusion oxygenation and ventilation with carbon dioxide in oxygen were studied and compared. On bleeding the dogs, there was a marked decrease in the blood flow to all regions of the kidney and the diuresis stopped. Re-transfusion or infusion of plasma expander increased the blood flow in all regions of the kidney and the anuria ceased.

ZUSAMMENFASSUNG

Es wurden die Veränderungen in der Nierendurchblutung – besonders der regionalen – bei normovolemischen und hypovolemischen Hunden studiert. Wärmeleitsonden im Anschluss an Gibbs wurden gebraucht. Die Wirkung von Adrenalin, Nor-Adrenalin, Methoxamin, Metaraminol, Normetaoxedrin, Arg^8-Vasopressin und Angiotensin II ist untersucht worden. Bei kontinuerlicher intravenöser Infusion von Nor-Adrenalin in Dosen bis zu 0,6 μg/kg · Minute nahm die Durchblutung des Marks zu. Gleichzetig

wuchs die Urinenproduktion an, während die Rindendurchblutung abnahm. Die übrigen Pressorsubstanzen ergaben geringere Durchblutung in sowohl der Rinde als auch dem Mark. Papaverin bewirkte, dass der arterielle Blutdruck durch generelle Vaso-Dilatation sank, wobei die Nierendurchblutung abnahm. Infusion von kleinen Dosen Bradykinin in die A.renalis ergab eine gesteigerte Rinden- und Markdurchblutung, ohne dass der Blutdruck dabei sank. Der Effekt von aponoeischer Diffusion–Oxygenation und Atmung mit einer Mischung von Kohlendioxyd und Sauerstoff wurde verglichen. Bei der Ausblutung nahm die Durchblutung in der Rinde und dem Mark stark ab, und die Urinenproduktion hörte auf. Retransfusion oder Infusion von Plasmaexpander steigerte die Durchblutung in der Rinde sowie im Mark, wobei die Anurie gehoben wurde.

INTRODUCTION

Previous studies, using PAH and Diodrast clearance, have shown that bleeding results in reduced renal blood flow (Corcoran and Page 1943, Lauson, Bradley and Cournand 1944). In recent years these results have been verified by direct flow measurements through an intact renal artery or renal vein, using an electromagnetic flow meter (Mills and Steppacher 1963, Fell 1966). The reduced renal blood flow is probably due to increased tone in the sympathetic nervous system, which primarily affects the skeletal musculature (Lundgren, Lundwall and Mellander 1964) and the splanchnicus area (Gernandt, Liljestrand and Zotterman 1946). By this means, vital organs, such as the brain and the heart, are directly assured of as good a blood flow as possible, at the expense of the blood flow to the liver, kidney and other organs. Normally the blood flow to the kidney is about 20% of the volume that flows thorugh the heart per minute, but in pronounced states of shock it may fall heavily. The impaired blood flow would seem to be critical, especially as regards the renal medulla. This is important, particularly as investigations in recent years (Kramer, Thurau and Deetjen 1960, Lilienfield and Maganzini 1963, Meier, Brechtelsbauer and Kramer 1964) have shown that the inner parts of the medulla and especially the papilla even under normal conditions have a very low blood flow, equivalent to one or a few per cent of the total renal blood flow. The low medullary blood flow has been verified by other research workers,

though they give somewhat higher absolute values (Thorburn 1963, Aukland 1964, Wolgast 1968). Besides the fact that the absolute blood flow to the medulla is relatively low, the marked counter-current exchange system will further restrict the nutritive capacity of the blood. In the renal medulla, oxygen will be shunted from descending to ascending vessels, while carbon dioxide and other metabolites formed in the medulla will be shunted from ascending to descending vessels, by which means carbon dioxide will be retained in the area. As far as the medulla and especially its inner parts are concerned, we have consequently to distinguish between the absolute blood flow (red cells and plasma) and the nutritive blood flow or effective blood flow.

Absolute blood flows are measured by using intravasal indicators, such as Evans blue (Kramer, Thurau and Deetjen 1960), albumin labelled with I^{131} (Lilienfield and Pomerantz 1963) and red cells labelled with P^{32} (Wolgast 1968), while the nutritive effect of the blood flow is studied by using the inert gas technique (Thorburn *et al.* 1963, Aukland and Berliner 1964) and heat-clearance methods (Scher 1951, Perl and Hirsch 1966, Grängsjö *et al.* 1966, Grängsjö 1968). By way of summary, we may say that none of the methods that have been worked out for measuring medullary blood flow is free from objection. This is due, to a large extent, to the complex circulatory conditions which exist in the kidney.

In the present investigation we used heated therocouples, as described by Gibbs (1933), to measure the nutritive flow, as a complement to the determinations of absolute flow. Heated thermocouples have the great advantage that with them it is possible to make a continuous record of the regional nutritive flow, any rapid and transitory changes of flow and any re-distributions of the blood flow between the cortex and the medulla. In addition, they are simple to use and the trauma to which the kidney is subjected in inserting them at the measuring site is very slight, compared with those caused by other probes for measuring the regional blood flow.

The purpose of our investigation was to study in the dog the effect on the renal blood flow, and especially the regional blood flow, of some vaso-active substances, carbon dioxide, bleeding and re-transfusion. The vaso-active substances we chose were, on the one hand, the pressor substances adrenaline, noradrenaline, methoxamine, metaraminol and normetaoxedrin (Effontil), together with Arg^8-Vasopressin (ADH) and angiotensin II, and, on the other, the dilatator substances papaverine and bradykinin. In addi-

tion, we recorded the flow changes due to the form of anaesthesia and the effect of increased p_{CO_2} in apnoeic diffusion oxygenation (Holmdahl 1956) and in ventilation with different concentrations of CO_2 in the inhalation gas. In bleeding the experimental animal, we studied exclusively the acute changes and the effect of the vaso-active substances in the acute phase, together with the significance of the restoration of the blood volume, by transfusing sometimes blood and sometimes plasma expander.

The changes in blood flow are presented qualitatively. A change in thermocouple response greater than one half of a per cent was considered as indicating a real change in nutrient blood flow.

METHODS

The investigation was carried out on a series of 45 mongrel dogs, which varied in weight between 6.5 and 35 kg. Up to the beginning of the experiment the dogs had free access to water but received no solid food for 12 hours before the inception of the anaesthesia. At the beginning of the series of experiments the dogs were anaesthetized with pentobarbital sodium (30 mg per kg body weight i.v.), but this form of anaesthesia proved to have many disadvantages, including cardiovascular effects (Olmsted and Page 1966) and the release of an antidiuretic hormone (Sawyer 1961, Papper and Papper 1964). We therefore changed to chloralose, which was administered until full anaesthesia was achieved (40–50 mg per kg body weight i.v.), usually after the induction of initial anaesthesia with thiopentone sodium★ (20 mg per kg body weight i.v.). Chloralose proved to have a slight effect on the cardiovascular system (Van Citters, Franklin and Rushmer 1964) and does not seem to produce any marked release of antidiuretic hormone (Grängsjö 1968). During the experiments the anaesthesia was maintained with pentobarbital sodium (30–50 mg per hour) or chloralose (200–300 mg per hour). No premedication was given. A free respiratory passage was ensured by intubation.

★ Thiopentone sodium (Pentothalsodium^R), was kindly placed at our disposal by Abbot AB, Stockholm, Sweden.

OPERATIONAL TECHNIQUE

Anaesthetics and drugs were administered through a polythene catheter inserted in a superficial vein in a leg. The mean arterial pressure was recorded via a catheter inserted in the femoral artery and connected to a pressure transducer. The dogs could be bled through this catheter, in order to bring about hypovolemic states. The left kidney was exposed extraperitoneally by a flank incision. In the experiments in which the total flow to the kidney was recorded, the renal artery was prepared free, after which it was connected with the common carotid artery or the femoral artery via a system of tubes maintained at a temperature of 37°C by a thermostat. In some experiments a rotameter (Shipley 1951) was connected in this system of tubes and sometimes also a Sigma pump, by which the blood flow to the kidney could be regulated. A "wind-kessel" was connected after the Sigma pump. In the cases in which the total flow was measured, the dogs were heparinized with 15–20 mg of heparin per kg body weight.★ Injections could be made through the system of tubes directly into the renal artery. The renal perfusion pressure was recorded by a pressure transducer. Ureteral catheters were inserted supravesicularly through a middle-line incision, which was then sutured. The urine secretion was measured by a drop recorder, which was electronically reset after each 30th drop. 30 drops had a volume of 1 ml. For continuous injections an injection apparatus with a variable speed was used.

HEATED THERMOCOUPLES

In the determinations of intrarenal flow we used heated thermocouples of the same design as those described in previous papers (Grängsjö *et al.* 1966, Grängsjö 1968) (Fig. 1). The thermocouple consisted of a 7–8–mm–long constantan wire, 0.10 mm in diameter, to the two ends of which were soldered an insulated copper wire, 0.10 mm in diameter. The constantan wire was divided by the copper wire BC, which was soldered to point M, into a short section (MS), 2–3 mm long, and a long section (MR), 4–5 mm long. The short section was heated by a stabilized AC current (50 c/s)

★ The heparin was kindly supplied by Vitrum AB, Stockholm, Sweden.

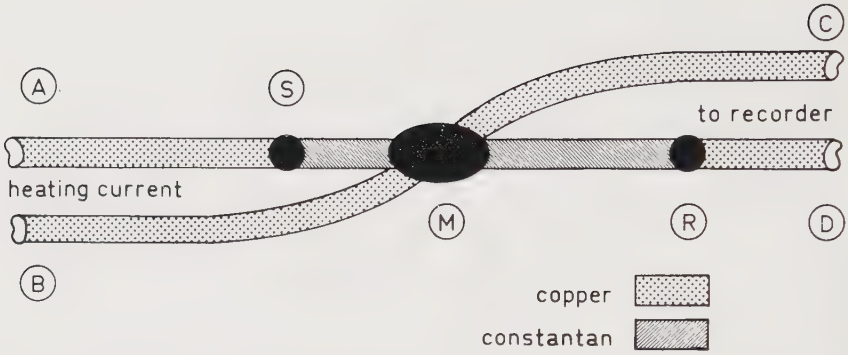

Fig. 1. The design of the heated thermocouple. MS is the heated section o the constantan wire and R the reference point, 3–4 mm away from M.

between A and B; which was chosen so that the renal parenchyma next to point M was heated maximally by about 2.5°C if there was no blood flow. The connecting wires for the heating section were soldered end–to–end to a needle, 0.25 mm in diameter, which was stuck through the kidney and thus the thermocouple could be drawn to the desired place in the parenchyma. Its exact placement was determined by careful dissection of a kidney after fixation for a day or two in 6% formalin solution.

The thermo-electric current due to the temperature difference between the measuring point, M, and the reference point, R, was recorded by a UV recorder. When the blood flow past the thermocouple was nil, this temperature difference was at its maximum and the position of the galvanometer pointer then formed our zero-line, from which the thermocouple response was measured. When the blood flow increases, the temperature difference decreases and the deviation from the zero-line is a measure of the ability of the blood flow to cool down the heated soldered point M (the nutritive effect of the blood flow). In a previous paper (Grängsjö *et al.* 1966) it was shown that the calibration curve for a thermocouple in homogeneously perfused tissue forms a hyperbola, if the deviation from the zeroline is plotted against the blood flow.

RESULTS

EFFECT OF ANAESTHETIC

Pentobarbital sodium was administered to 14 of the 45 dogs in the series until full anaesthesia was achieved. On the injection of the maintenance doses a fall in blood pressure of 10–20 mm Hg was always recorded, but the original pressure was restored after 2–4 minutes. The regional flow decreased at all levels in the kidney but reached the initial value at the earliest 5 min. after the blood pressure had been restored. Similarly, the diuresis decreased considerably, often to less than half, and did not reach the initial value until the blood flow had been "normalized".

On the other hand, chloralose, which was used on 31 dogs, did not produce any recordable changes in the blood pressure, regional flow or diuresis, unless very large quantities were injected rapidly or the temperature of the solution differed a good deal from the temperature of the dog's body. In protracted experiments we sometimes noted haematuria, which was possibly due to the chloralose anaesthesia, as we did not find it with other forms of anaesthetic.

Nitrous oxide, 80% in oxygen, did not seem to effect either the blood flow or the diuresis.

Succinylcholine produced a slightly reduced blood flow in both the cortex and the medulla and a reduced diuresis, in spite of an unchanged or only slightly lowered blood pressure.

VASOPRESSOR SUBSTANCES

Adrenaline. The series comprises 10 normovolemic dogs, with mean arterial blood pressures between 110 and 130 mm Hg. The adrenaline was administered as single doses and as continuous injections, both intravenously and intra-arterially in the renal artery. The doses varied between 0.13 and 21 μg per kg for the single injections and between 0.06 and 5.7 μg/kg · min. for the continuous injections. There was no difference in effect between intra-arterial and intravenous infusion and therefore they are reported together in Fig. 2. The thermocouples recorded a reduced nutritive flow in both cortex and medulla. In the inner zone we found an increase in flow

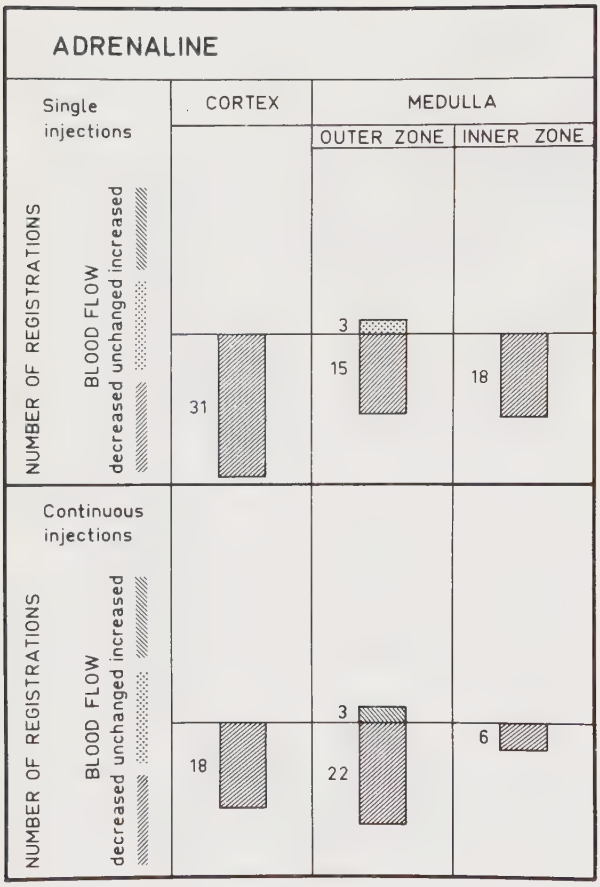

Fig. 2. Summary of the regional blood flow changes, with single and continuous injections of adrenaline. The material includes both intra-arterial and intravenous injections. The series comprises 10 normovolemic dogs.

initially but afterwards the flow decreased and the net result was always a reduced flow (Fig. 3). The reduction in flow recorded by the thermocouples was greater for greater doses of adrenaline, which agreed with the direct measurements by rotameter in the renal artery. The systemic blood pressure and perfusion pressure showed greater increases for greater doses. The diuresis always decreased and fell to zero for larger doses. Often but not regularly, we were able to record an excess in the flow, when the adrenaline effect had passed over.

Noradrenaline. The series comprises 11 dogs, four of which were in states of haemorrhagic shock, produced by drawing 400–600 ml of blood from them, until the mean blood pressure had fallen to about 80 mm Hg. Noradrenaline was administered intravenously, partly as a single injection and partly as a continuous infusion into both normovolemic and hypo-

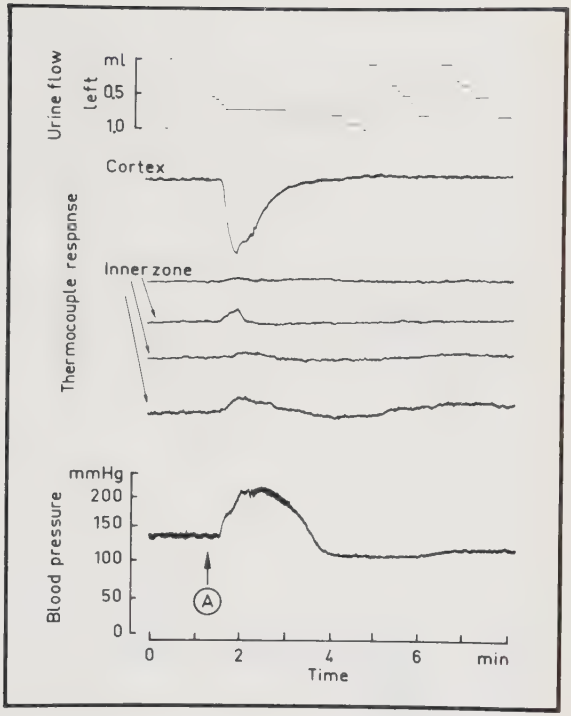

Fig. 3. The effect of a single intravenous injection of adrenaline (1.2 μg/kg) at A. The blood pressure increases greatly, the cortical blood flow decreases and the diuresis ceases for 3–4 minutes. The blood flow in the inner zone increases initially but then decreases.

volemic dogs. The single injections varied between 0.8 and 8 μg per kg and the continuous injections between 0.03 and 22.5 μg/kg · min. The results are shown in Figs. 4 and 5.

With *the single injections,* a reduced blood flow was recorded in both normovolemic and hypovolemic dogs in the renal cortex and medulla. The systemic blood pressure showed greater increases with greater doses. With the small doses the diuresis was unchanged, but with the greater doses the flow of urine decreased.

With *the continuous injections* of noradrenaline, we found a different pattern. In both normovolemic and hypovolemic dogs, intravenous infusions of up to about 0.2 μg/kg · min. produced no change of blood pressure, regional blood flow or diuresis. Increased doses produced a *reduced* blood flow in the cortex but an *increased* blood flow in the outer and inner zones of the medulla, together with an *increased* flow of urine, with doses of up to about 0.6 μg/kg · min., while the blood pressure increased slightly. In the normovolemic dogs the blood pressure increased to a maximum of 145 mm Hg and in the hypovolemic dogs from about 80 to about 110 mm Hg. With a further increase of the speed of infusion, the blood flow decreased

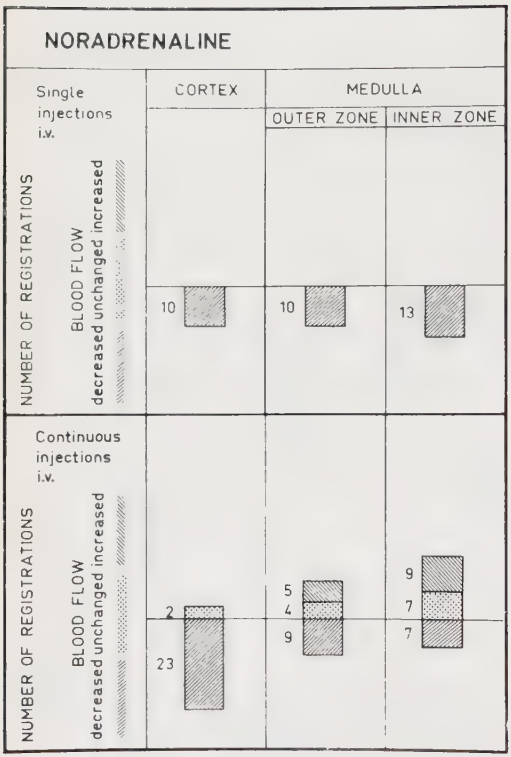

Fig. 4

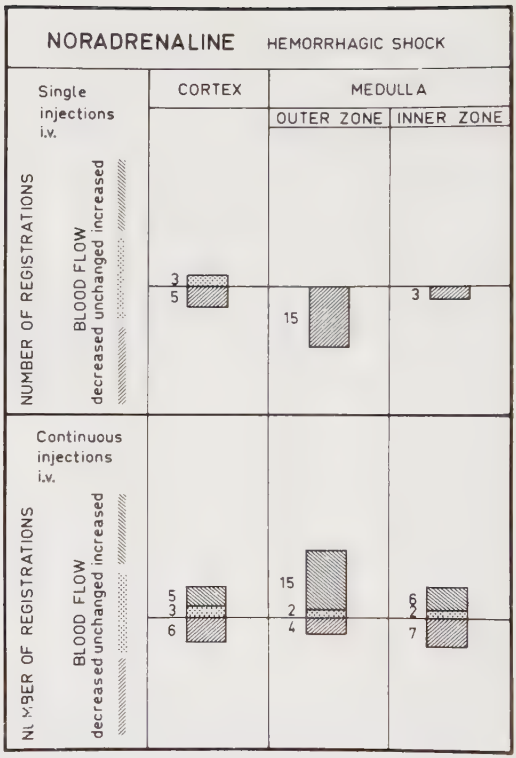

Fig. 5

Fig. 4. Summary of the regional blood-flow changes, with intravenous injections of noradrenaline. Single injections resulted in reduced blood flows in both cortex and medulla. Continuous injections in doses <0.2 μg/kg · min. produced no changes, while doses of 0.2–0.6 μg/kg · min. reduced the cortical blood flow but increased the medullary blood flow. In doses >0.6 μg/kg · min. the blood flow decreased in all regions. The series comprises 10 normovolemic dogs.

Fig. 5. Summary of the regional blood-flow changes, with intravenous injections of noradrenaline into hypovolemic dogs. Single injections resulted in reduced blood flows in both cortex and medulla. With continuous injection (0.2–0.6 μg/kg · min.), the flow increased in all regions, while the diuresis increased. The series comprises four dogs.

in all the regions of the kidney and the diuresis decreased, in spite of a further rise in the systemic blood pressure.

Methoxamine chloride (Vasoxine R, Burroughs Wellcome, England) was administered i.v. to three dogs with normal blood pressure (110–120 mm Hg). The doses varied between 0.17 and 0.6 mg per kg body weight. The blood pressure increased to maximally 140 mm Hg. The diuresis decreased substantially and in the first few minutes after the infusion it ceased entirely. In one dog in which a rotameter was connected in the renal artery, the flow fell from 110 to 10 ml per min. At the same time the systemic blood pressure increased from 110 to 140 mm Hg. Regional blood flows were recorded only in the cortex and only in two dogs, but greatly reduced flows were noted in these cases.

In three dogs in hypotension the effect of intravenous doses from 0.3 to

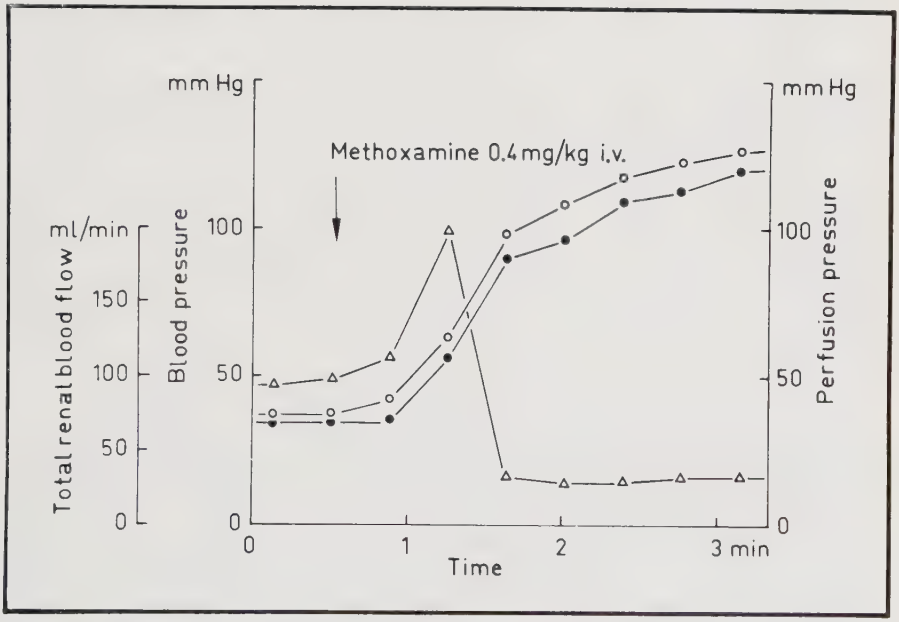

Fig. 6. The effect of a single intravenous injection of methoxamine on a dog in a state of haemorrhagic shock. The open circles represent the arterial blood pressure, the solid circles the perfusion pressure in the renal artery and the open triangles the blood flow in the renal artery, measured with a rotameter. After an initial increase, the blood flow decreases substanitally, in spite of the fact that the systemic blood pressure and the perfusion pressure continue to increase.

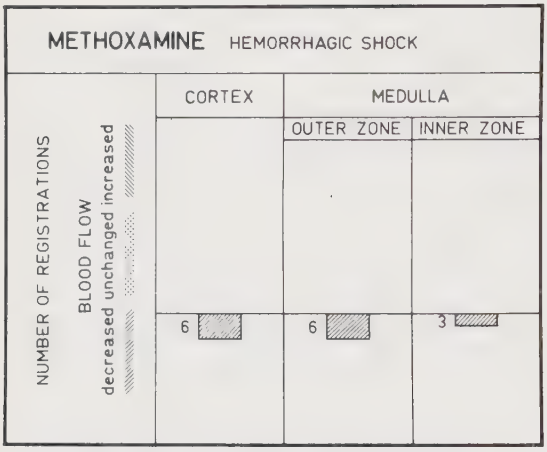

Fig. 7. Summary of the regional blood-flow changes, with single intravenous injections of methoxamine into three dogs in states of haemorrhagic shock.

1 mg per kg body weight was studied. The systemic blood pressure increased from 110 to 155 mm Hg. The blood flow in the renal artery fell heavily to 20–30% of the original value (Fig. 6). We recorded with the thermocouples a substantially reduced nutritive flow in all regions (Fig. 7). As the effect of methoxamine remains for at least an hour, there was no reason to study the effect of continuous injections.

Metaraminol (Aramine^{R})*, was administered to one dog in the form of single injections in the renal artery (0.6 – 1.3 μg/kg). After bleeding the dog to a blood pressure of 65 mm Hg, single intravenous injections ranging from 6 to 312 μg per kg body weight were given. As this preparation is a long-acting one, it was considered unnecessary to study the effect of continuous injections. The intra-arterial injections produced in 5–10 min. a substantially reduced flow in the renal artery, measured by a rotameter, together with a decrease in the diuresis which remained for 20–30 min. No effect on the systemic blood pressure was recorded. The intravenous injections reduced the flow in the renal artery and decreased the flow of urine during the time for which the increase in blood pressure subsisted (Fig. 8). Reduced flows in both the cortex and the medulla were recorded by the thermocouples. The greater the dose of metaraminol, the greater the increase in blood pressure and the greater the reduction in the flow of blood to the kidney.

N-ethyl-normetaoxedrin (Effontil ^{R})**, was administered to a dog in a state of hypovolemic shock in the form of single injections intravenously

* The Aramine was kindly placed at our disposal by MSD, New York, U.S.A.

** The Effontil was kindly placed at our disposal by Böhringer/Ingelheim, Germany.

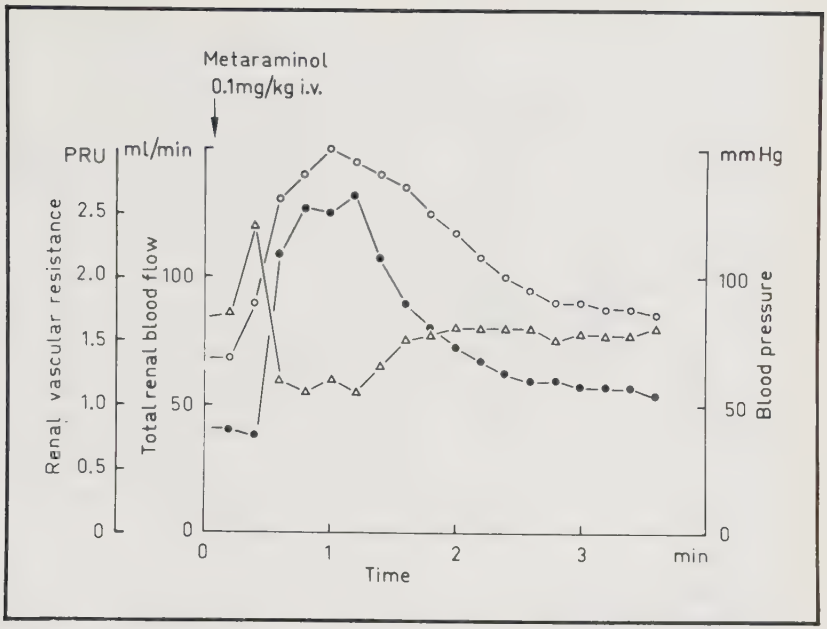

Fig. 8. The effect of a single intravenous injection of metaraminol. The open circles represent the blood pressure, the solid circles the renal vascular resistance (PRU) expressed in mm Hg · min. · ml^{-1}, and the open triangles the blood flow in the renal artery. The blood pressure increases, but the resistance increases more vigorously and the net effect is a reduced renal blood flow.

(10 and 20 μg per kg body weight). The results were identical with those produced by metaraminol and methoxamine, namely, increased blood pressure but decreased diuresis and decreased nutritive flow in the cortex and medulla as long as the blood-pressure-raising effect subsisted.

Arg^8-Vasopressin,★★★ the anti-diuretic hormone in the dog, was administered to a series of six dogs in continuous injections intravenously ranging from 0.0017 to 0.16 IU/kg · min. The experiments,showed that low concentrations did not produce any effect on the blood pressure. However, the osmolality of the urine increased and the diuresis decreased. On the other hand, the regional blood flow in the kidney, measured by thermocouples, was unchanged. At higher concentrations the blood pressure rose, but at the same time the thermocouple responses in both cortex and medul-

★★★ The Arg^8-Vasopressin was kindly placed at our disposal by Ferring AB, Malmö, Sweden.

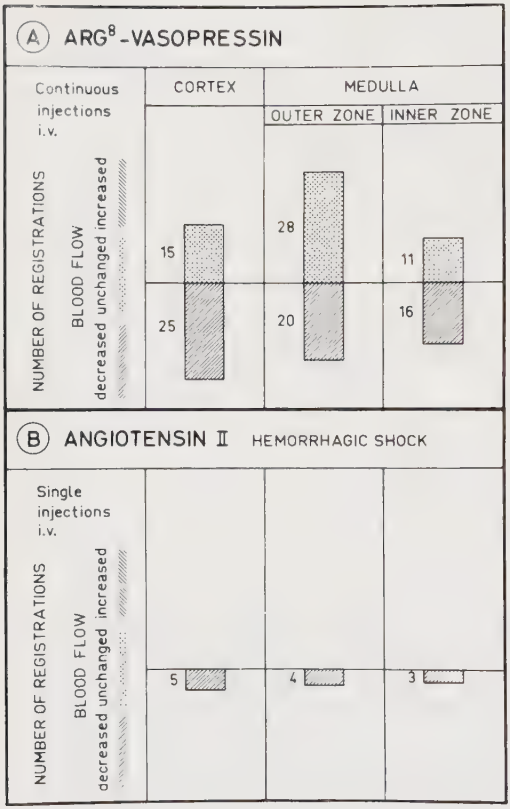

Fig. 9. Summary of regional blood-flow changes in the kidney. A. Continuous intravenous injections of Arg^8-Vasopressin (ADH). In physiological blood concentrations, ADH produces no change in the blood flow. In greater concentrations, a decreased flow is obtained in both cortex and medulla. B. Single intravenous injections of angiotensin II into hypovolemic dogs resulted in reduced blood flows in both cortex and medulla.

la decreased, which was interpreted as indicating reduced nutritive flow. A summary of the results is shown in Fig. 9 A. The effect of the anti-diuretic hormone in hypotension was not studied.

Angiotensin II (Hypertensin N^R) ★ was administered in the form of single intravenous injections in one normovolemic dog with normal blood pressure and in three hypovolemic dogs whose blood pressures were initially between 75 and 80 mm Hg. The doses varied between 2 and 23 mg per kg. The results, are interpreted as meaning that angiotensin produces a greatly reduced nutritive flow in both the cortex and the medulla in both normovolemic and hypovolemic dogs. A summary of the results is shown in Fig. 9 B. With the low doses the blood pressure increased to 140–150 mm Hg and with the higher doses to 180–200 mm Hg. The diuresis decreased substantially and ceased entirely during the first few minutes after the injection.

★ The Hypertensin N was kindly placed at our disposal by Ciba, Basle, Switzerland.

VASODILATING SUBSTANCES

Papaverine was given to eight dogs in single doses and in continuous injections (0.12 – 4 mg/kg and 0.8 – 4.3 mg/kg · min. respectively). It decreased the resistance in the renal vessels but at the same time lowered the systemic blood pressure. The net effect of papaverine, as far as the kidney was concerned, was a decreased blood flow in the renal artery (measured by a rotameter) and a reduced regional blood flow in the cortex and medulla (measured by thermocouples), together with decreased diuresis. The resistance in the renal vessels decreased, with the larger doses by 4–5 times, and in the cases in which we kept the perfusion pressure in the renal artery at a normal level with a Sigma pump, the blood flow increased greatly in all regions and the diuresis increased.

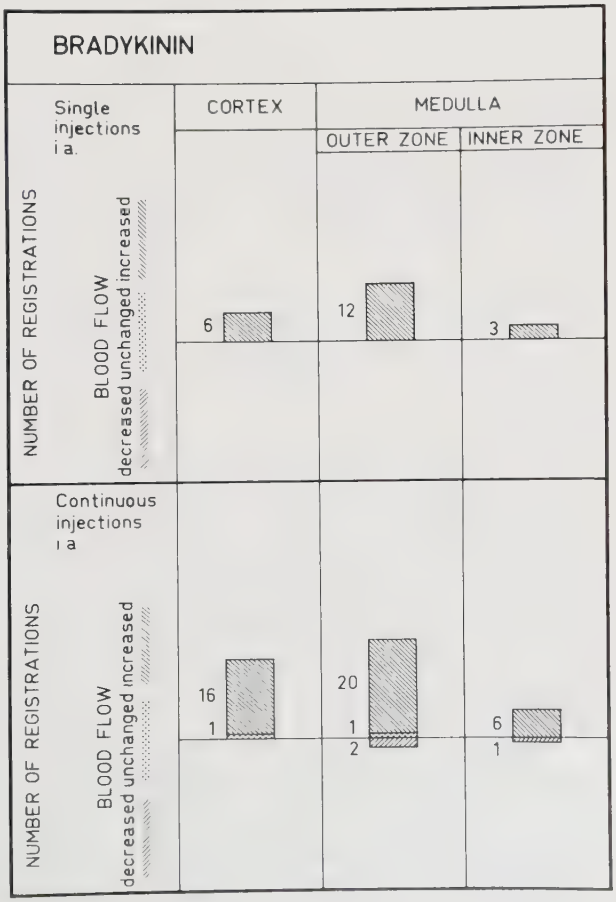

Fig. 10. Summary of effects of single and continuous injections of bradykinin in the renal artery. Blood-flow increases were recorded in both cortex and medulla, with doses which did not affect the systemic blood pressure.

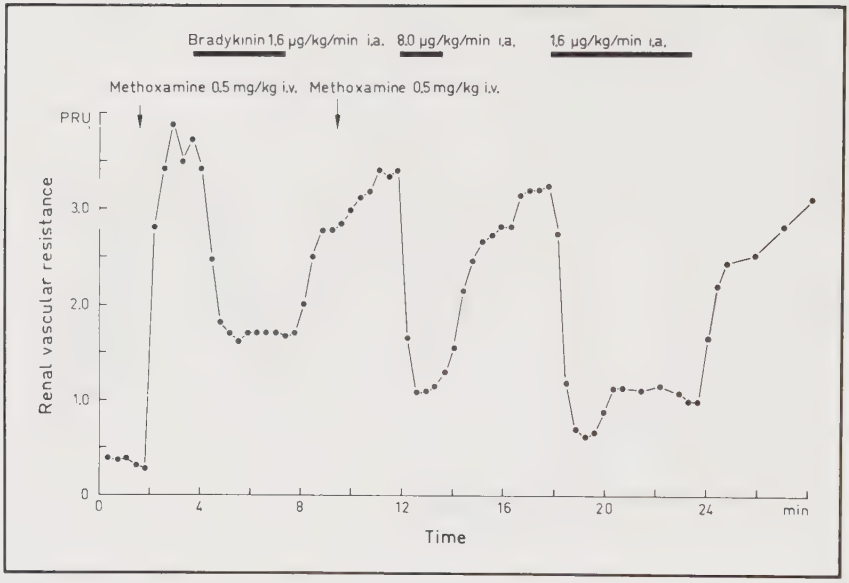

Fig. 11. This figure shows how the vascular resistance (mm Hg · min. : ml in the kidney increases by 8–10 times after the injection of methoxamine. However, the effect may be largely cancelled by the continuous intra-arter infusion of bradykinin.

Bradykinin ★ was studied in a series of five dogs. As bradykinin has a very short biological half time (20–30 seconds), it has to be administered intra-arterially. Intravenous injections of a volume great enough for a sufficient quantity to reach the kidney and have a vasodilating effect produced a general vasodilatation and thereby brought about a reduced system blood pressure and a reduced flow of blood to the kidney, though the vascular resistance in the kidney decreased by 2–3 times. The intra-arterial injections were given partly as single doses (1–8 µg/kg) and partly as continuous injections (0.04–8 µg/kg · min.). The results, which are reported in Fig. 10, have a very homogeneous pattern, with increased blood flow in all regions of the kidney. Increased blood flows in the renal artery were recorded in the cases in which the rotameter was connected. At the same time the flow of urine increased and the urine became isotonic or even slightly hypotonic (270 mosm/kg). Fig. 11 shows how the renal vascular resis increased by about eight times after the injection of methoxamine and how

★ The bradykinin was kindly placed at our disposal by Sandoz AG, Basle, Switzerland.

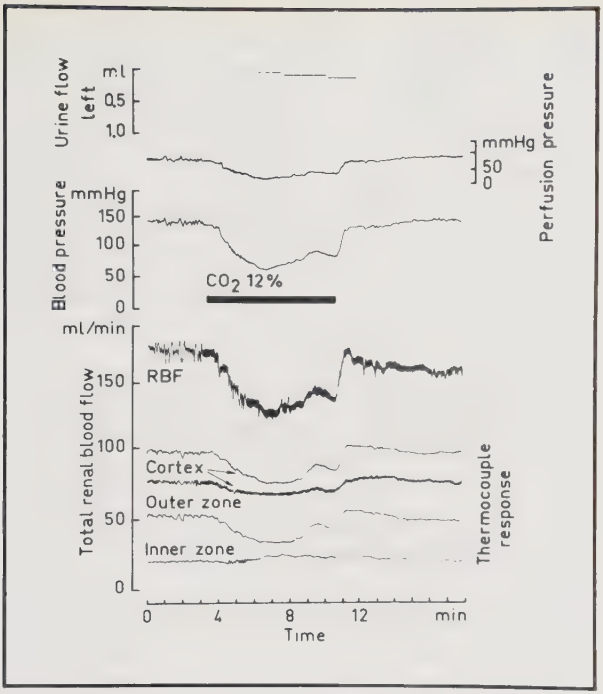

Fig. 12. This figure shows how the systemic blood pressure and the perfusion pressure in the renal artery decrease on ventilation with 12% carbon dioxide and how the urine production almost ceases. The thermocouples record substantially reduced flows in the cortex and the outer zone. In the inner zone, on the other hand, a slightly increased flow is recorded. The blood flow in the renal artery (RBF) decreases substantially but increases when the carbon dioxide is removed.

this drastic vascular constriction could be abolished by bradykinin. Bradykinin also had the same effect on other occasions, producing increased resistance in the renal vessels.

Effect of CO_2. The effect of ventilation with different concentrations of CO_2 in oxygen was studied in five dogs. The concentrations varied between 5% and 20% CO_2. In the experiments, a *lowered* blood pressure, a decreased production of urine and a reduced nutritive flow were recorded in the different regions of the kidney (Fig. 12). In the inner zone the flow often increased, to begin with, but afterwards fell below the initial value. In the cases in which a rotameter was connected, a decreased flow was measured in the renal artery. However, the vasoconstriction in the renal vessels on account of the very high concentration of CO_2 in the inhalation

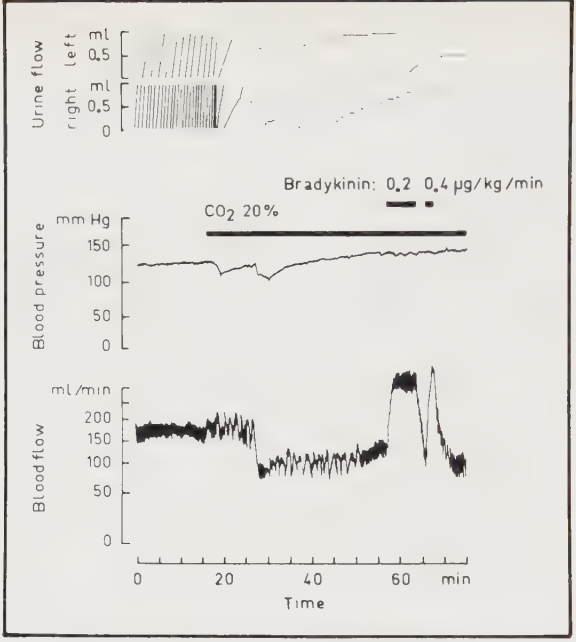

Fig. 13. This figure shows how the blood flow in the left renal artery decreases when the dog is ventilated with 20% carbon dioxide. The urine production decreases substantially. Bradykinin infused into the left renal artery increases the blood flow. The diuresis on the left side increases, while on the right side it decreases further. The blood pressure falls, to begin with, and then rises to slightly above the initial value.

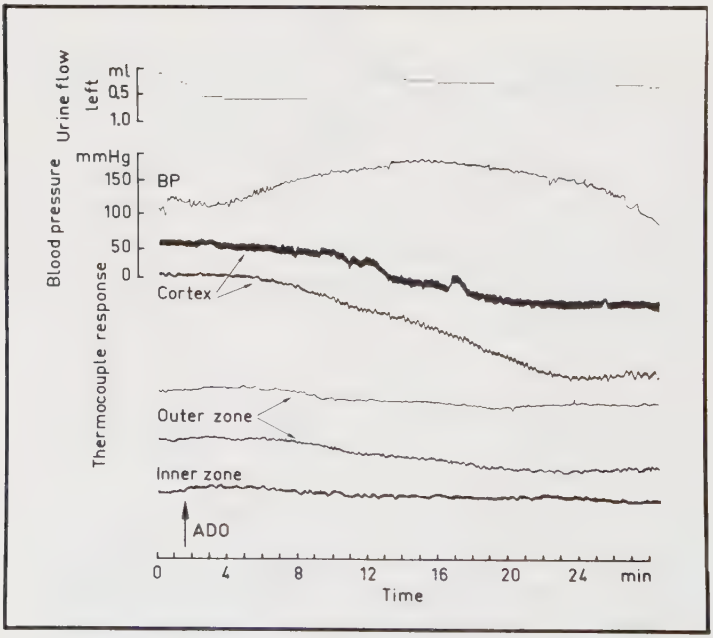

Fig. 14. Apnoëic diffusion oxygenation (ADO), in which the blood pressure rises and the diuresis ceases almost entirely. The thermocouples record substantially reduced flows in all regions.

gas (20%) could be abolished by injecting bradykinin continuously into the renal artery. The blood flow to the kidney increased and the diuresis began (Fig. 13).

For comparison, we studied in one dog the flow changes which arose in the kidney in the course of apnoeic diffusion oxygenation (Fig. 14). After

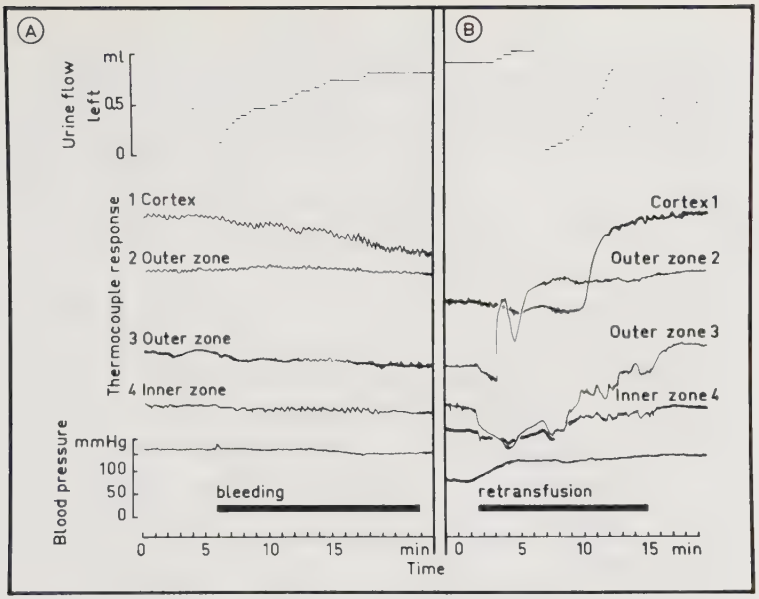

Fig. 15. On bleeding the dog, (A) the blood flow decreases substanitally in all regions and the diuresis stops, in spite of the fact that the blood pressure is still unchanged eight minutes after the bleeding has begun and that 200 ml (corresponding to about 15% of the dog's total blood volume) has been drawn. In all, 500 ml of blood were drawn from the dog and were re-transfused (B) about one hour later. The blood flow increases greatly in the cortex and medulla, the blood pressure is normalized and the production of urine begins.

the dog had been made to breathe 100% oxygen for 36 minutes, a neuromuscular blockade was laid down with succinylcholine, 0.6 mg/kg, which resulted in a respiratory standstill for more than 30 minutes. During the first 15 minutes *the blood pressure increased,* the urine production ceased and the nutritive flow in the renal cortex and medulla decreased very greatly. Thus, there was a clear difference in the effect of CO_2 retention through apnoeic diffusion oxygenation and through spontaneous respiration with CO_2 in the air inhaled.

Bleeding, even of small amounts, immediately initiated a heavy reduction of the renal blood flow in all regions, in spite of the fact that the systemic blood pressure remained at the original level. The urine production ceased almost immediately (Fig. 15 A). As has previously been mentioned, it was

possible to increase the systemic blood pressure by injecting various pressor substances. However, this resulted only in a further reduction of the absolute blood flow to the kidney, measured by a rotameter, and of the nutritive flow, both in the cortex and the medulla, as measured with heated thermocouples. It has already been mentioned that the only exception was noradrenaline, small doses of which were able to increase slightly the nutritive blood flow and the diuresis (Figs. 4 and 5).

Re-transfusion of the blood drawn off or the supply of plasma expander (Fig. 15 B) always had a very rapid and prompt effect. The systemic blood pressure increased, and also the renal blood flow. All the thermocouples showed a substantially increased nutritive flow and the diuresis, which had been entirely suspended during the period for which the hypovolemic shock lasted, started almost immediately. The effects of bleeding and re-transfusion were studied in a series of 10 dogs.

DISCUSSION

In the present investigation we studied in a series of altoghether 45 dogs the haemodynamic conditions in the kidney and the changes in it due to the supply of vaso-active substances and the effect of CO_2 and the acute changes which ensue as a result of haemorrhagic shock and re-transfusion.

ANAESTHESIA

To begin with, we anaesthetized the dogs with pentobarbital sodium but changed over to chloralose, which produced the least effects on the haemodynamics and the cardiovascular system (Smith 1951, Papper and Papper 1964, van Citters, Franklin and Rushmer 1964).

Experiments showed that nitrous oxide had little effect on the blood flow through the kidney, but this form of anaesthesia had to be combined with a neuromuscular blockade and artifical ventilation, which both produce great changes in the haemodynamics. Succinylcholine has been shown (Granberg and Wåhlin 1962) to reduce both the glomerulus filtration (inu-

in clearance) and the total flow to the kidney (PAH clearance), which agrees with the reduced regional blood flow in the cortex and medulla recorded in our experiments. In addition, we found a reduction in the flow of urine, a parameter which Granberg and Wåhlin could not study on account of their experimental conditions.

VASOPRESSOR SUBSTANCES

Sympathomimetic drugs. The effects of sympathomimetic drugs on the cardiovascular system as a whole and on the renal haemodynamics have been studied by many authors and the literature on this subject is particularly extensive. The statements made as to the effects of the different pressor amines are often at variance with each other and "the field is so complicated that research pharmacologists will be kept happy arguing with each other for many years" (Burton 1966). The intrarenal blood flow in haemorrhagic shock and the effects of vaso-active substances, on the other hand, have as yet been very little studied and the literature is relatively scanty and easy to survey.

PRESSOR AMINES

According to our investigations, *adrenaline, methoxamine, metaraminol* and *normetaoxedrin* decrease the blood flow in the cortex and medulla in both normovolemic and hypovolemic dogs. The increase in vascular resistance in the kidney was clearly so great that the net effect of the increased blood pressure and the, in most cases, increased cardiac output was a decreased renal blood flow. These results agree with those of Scher (1951), who recorded by heated thermocouples a reduced blood flow in the cortex and medulla after single injections of adrenaline and noradrenaline. They also agree with the results of the majority of other investigations in which a reduced blood flow in the renal artery was found on administration of these sympathomimetic drugs. The increase of p_{O_2} in the urine after the administration of small single doses of adrenaline which was found by Aukland and Krog (1961) and Ulfendahl (1962) may perhaps be explained by the transient increase of nutritive flow which we found in the inner zone

of the medulla and which is reported in Fig. 3. The excess which was often recorded in connection with the passing of the pressor effect may be interpreted as showing that reactive hyperaemia occurs in the kidney. However, its occurrence is disputed by other authors (*inter alia,* Grupp and Heimpel 1958).

Noradrenaline, administered by continuous injection intravenously in doses ranging from 0.2 to 0.6 μg/kg · min. produced, on the other hand, an *increased nutritive flow* in the cortex and medulla, together with *increased diuresis,* in both normotensive and hypotensive dogs. Greater doses produced a reduced blood flow and a reduced diuresis, in spite of a further rise in the mean blood pressure. These results agree with those of Langston *et al.* (1962), who , on infusing up to 0.6 μg/kg · min. of noradrenaline into a dog, found an increase of the renal blood flow, the glomerulus filtration and the diuresis. Mills (1965) also found an increase of 3.6% in the blood flow in the renal artery following the intravenous infusion of noradrenaline in doses which produced a 15% increase in the arterial pressure.

PRESSOR PEPTIDES

Arg^8-Vasopressin, the specific antidiuretic hormone in the dog, had no effect on the regional renal blood flow when continuously infused in physiological doses but reduced the diuresis and increased the urine osmolality. Aukland (1967) draws the same conclusions from the results of measuring the hydrogen clearance in the outer zone of the medulla. Kramer *et al.* (cited in Thurau 1964) were also unable to demonstrate any vaso-active effect of ADH in physiological concentrations on the medullary vessels.

Angiotensin II produced a marked decrease in the nutrient blood flow to the renal cortex and medulla, in both normotensive and hypotensive dogs. No other authors have studied the intrarenal flow changes caused by angiotensin II, but the results agree well with the substantial reduction of the blood flow in the renal artery observed by many authors (*inter alia,* Peart 1959, Udhoji, Weil and Sambhi 1964, and Mills 1965). The reason for this substantial reduction of flow, combined with almost complete anuria, is probably a marked vasoconstriction on the arterial side (Cohn 1964).

The angiotensin II we used contained some mannitol, but the quantity was so small that it was certainly of no importance, as regarded the haemodynamics in the kidney.

VASODILATING SUBSTANCES

Papaverine produced general vasodilatation, substantially reduced blood pressure and decreased renal blood flow as net results (in spite of the greatly lowered renal vascular resistance), together with reduced regional blood flow and decreased diuresis. The results are in contrast to those of Harsing *et al.* (1964), who found, on injecting altogether 200 mg of papaverine for 10 min. into dogs anaesthetized with chloralose, that there was an increased renal blood flow, measured in the renal vein, and an increased flow of urine but only a small reduction in the mean arterial blood pressure. With equivalent doses of papaverine, we obtained general vasodilatation of such an order that the dog died, a difference in the effect of papaverine which is difficult to explain.

Bradykinin, infused continuously into the renal artery, produced increased renal blood flow in both the cortex and the medulla and increased the diuresis. Increased blood flow in the renal artery following continuous intra-arterial injection was reported in 1964 (Grängsjö, Ulfendahl and Wolgast 1966) and has also been confirmed by other workers (Goldberg, Dollery and Pentecost 1965, Furuyama *et al.*1966). Arcidiacono, Reiniger and Sapirstein (1967) found that intravenous injection of bradykinin had no effect on the renal blood flow, probably on account of the fact that bradykinin is rapidly decomposed and that none or only a very small part of the dose reached the kidney. The decreased osmolality of the urine is an interesting observation. This may perhaps be explained by saying that the hypertonic environment created in the papilla by the counter-current system is "washed away" by the increased medullary blood flow and the increased diuresis. Competition between the bradykinin and the antidiuretic hormone is not entirely excluded, as both the substances are peptides containing nine amino acids, several of which are common to both substances. Bradykinin also has an effect on biological membrane and increases the permeability, as was first shown by Holdstock (cited in Collier 1968). When infused intra-arterially, bradykinin produces dilatation of arterioles and capillaries (Hyman and Paldino 1967).

Bradykinin seems to be a substance which may come into great clinical use, for example, in transplantation surgery, for the improvement of the blood flow to individual organs or parts of the body. However, one disadvantage is that it has to be administered intra-arterially.

INFLUENCE OF CO_2

Whether it was due to ventilation with 5–20% of CO_2 in oxygen or to apnoeic diffusion oxygenation (Holmdahl 1956), hypercapnia resulted in a reduction of the nutritive flow in the renal cortex and medulla. However, the flow in the inner zone increased transiently – a re-distribution of the renal blood flow, possibly caused by the slower reactivity of the medulla.

The arterial blood pressure decreased on ventilation with CO_2, which is somewhat puzzling with regard to the powerfully stimulating effect which CO_2 has on the vasomotor centre (Guyton 1966). Under our experimental conditions, however, we obtained a reduced arterial blood pressure with a reduced renal blood flow as a result. Maybe that the reduced blood pressure was due to the depth of anaesthesia. The increase in blood pressure which we obtained in apnoeic diffusion oxygenation is in agreement with the results of previous investigations (Holmdahl 1956). The reduced renal blood flow is no doubt due partly to decreased cardiac output and partly to a general vasoconstriction, possibly caused by reflex mechanisms released by receptors in the lungs – a reflex of the same nature or identical with the diving reflex (Holmdahl and Schollander 1968).

HAEMORRHAGE AND RE-TRANSFUSION

In our experiments with hypovolemic dogs we had no intention of creating a state of standardized shock and only wished to study the acute changes in the renal blood flow caused by bleeding.

Carriere *et al.* (1966) interpreted their Kr^{85} "wash-out" curves as meaning that the cortical blood flow decreased but that the medullary blood flow usually increased in haemorrhagic hypotension. Truniger *et al.* (1966) interpreted Xe^{133} "wash-out" curves as meaning that the cortical flow decreased but that the medullary flow remained unchanged. On the other hand, we found a substantial reduction of the nutritive flow, both in the cortex and the medulla, and these findings are in agreement with those of Kramer *et al.* (1962) and Aukland and Wolgast (1968).

Re-transfusion or infusion of plasma expander resulted in increased flow in the renal artery and increased nutritive flow in the cortex and in the outer and inner zones of the medulla, and at the same time the existing

anuria was abolished. These results are in agreement with those of the majority of other authors (for example, Haddy *et al.* 1965).

By way of summary, we may say that in cases of haemorrhagic shock the blood-pressure-increasing drugs should only be used in emergencies and for a short time, in order to ensure the flow of blood to the immediate vital organs. On the other hand, with the available pressor substances, it seems impossible to increase the blood flow to the kidneys and thereby prevent acute renal failure.

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AUTOREGULATION OF REGIONAL BLOOD FLOW IN THE DOG KIDNEY

by

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UPPSALA 1968

ABSTRACT

Grängsjö, G., Autoregulation of regional blood flow in the dog kidney.

Autoregulation of the total renal blood flow was found in dogs anaesthetized with pentobarbital or chloralose, but could be eliminated by papaverine or bradykinin. The regional blood flow was studied with heated thermocouples. The response from thermocouples located in the outer or inner zone of the medulla were plotted against the perfusion pressure created by a Sigma pump. This resulted in S-shaped curves, which were interpreted as indicating the existence of autoregulation of the blood flow to the medulla. Further on graded constriction of the extracorporeal tube system, through which the left kidney was perfused, autoregulation was found in the renal cortex as in the outer and inner zones of the medulla. Sometimes, however, increased thermocouple response was obtained from the inner zone at perfusion pressures of 40–60 mm Hg. These findings may be interpreted as a redistribution of the renal blood flow, with increased medullary blood flow on hypotension.

Introduction

The existence of autoregulation of the total renal blood flow is well established. This was discovered by Rein (1931) and since then many authors have been occupied with the problems connected with auto-regulation, but the mechanism or mechanisms are still uncertain.

The counter-current hypothesis, and its dependence upon the medullary blood flow, in particular, has also greatly increased the interest in the distribution of the intrarenal blood flow. There is general agreement that the blood flow to the cortex, which constitutes about 80% of the total flow in the renal artery, is subject to autoregulation. Opinions are divided, however, as to the presence of autoregulation in the medulla. Only a few investigators, however, have studied the pressure-flow relationship with respect to the medullary blood flow.

From analyses of indicator dilution curves, Thurau et al. (1960) concluded that autoregulation did not occur in the medulla. This opinion has also been shared by Moffat (1967). Aukland (1967), on the other hand, on measurement of hydrogen gas clearance, found a pronounced autoregulation of the blood flow in the outer zone of the medulla. Autoregulation in the medulla has also been found by Wolgast (1968) on analysis of indicator dilution curves, which were recorded by small, beta-sensitive semiconductors placed at different levels in the kidney.

The aim of this investigation was to study the autoregulation of the kidney and the way in which this was influenced by papaverine and bradykinin, and also, by means of heated thermocouples placed in different regions of the kidney, to attempt to determine the blood flow changes caused by variations in perfusion pressure, measured in the renal artery.

EXPERIMENTS

Methods

The investigation was carried out on a series of 10 mongrel dogs, varying in weight between 10.5 and 23 kg. The dogs were fasted for 12 hours before the beginning of the experiments, but had free access to water up to

the time of anaesthesia. In six of the dogs anaesthesia was induced with i.v. pentobarbital sodium in a dose of 30 mg/kg body weight, and a maintenance dose of 30–50 mg/hour was then given. In four dogs anaesthesia was induced with i.v. thiopentone sodium* in a dose of 20 mg/kg body weight, after which chloralose in a dose of 40–50 mg/kg body weight was given until full anaesthesia was obtained. This was followed by a maintenance dose of chloralose of 200–300 mg/hour, which as in the case of the pentobarbital sodium was given through a polythene catheter introduced into a superficial vein in the leg. No premedication was given. All dogs were intubated immediately after induction of the anaesthesia.

The mean arterial blood pressure was recorded via a catheter inserted in the femoral artery and connected to a pressure transducer. The left kidney was exposed through a flank incision and its arteries were dissected free, care being taken to damage the renal nerves as little as possible. The left renal artery was cannulated with a polythene tube and connected to the proximal end of one or both carotid arteries. This extracorporeal connection was maintained at 37°C by a thermostatically controlled water jacket. The blood flow to the left kidney was recorded by means of a recording rotameter (Shipley and Wilson, 1951) and could be decreased by clamping or changed by means of a Sigma pump, after which a "wind-kessel" was connected. The dogs were heparinized with a dose of 15–20 mg/kg body weight.** Injections could be given directly into the renal artery through the tube system. The pressure at which the kidney was perfused was recorded via a pressure transducer.

The urine from the left kidney was collected by means of a catheter introduced into the ureter, through the flank incision, and the urine flow recorded by a drop counter.

Heated thermocouples were used for measuring the regional blood flow in the kidney, and were of the same construction as those described in previous papers (Grängsjö et al. 1966, Grängsjö 1968).

Papaverine when used was given intravenously. Bradykinin had to be given directly into the renal artery as continuous injections, because of its short biological half time of 20–30 seconds.

* Thiopentone sodium was kindly supplied by Abbot AB, Stockholm, Sweden.

** Heparin was kindly supplied by Vitrum AB, Stockholm, Sweden.

RESULTS

Autoregulation of total flow

On regulation of the blood flow in the renal artery with a Sigma pump, autoregulation was found both in dogs anaesthetized with pentobarbital (Fig. 1) and in those anaesthetized with chloralose (Fig. 2 and 4), and extended approximately over the range 90–220 mm Hg. By means of continuous i.v. injection of papaverine in a dose of 0.8 mg/min · kg body weight, the autoregulation could be eliminated, and the blood flow in the renal artery increased proportional to the increase in perfusion pressure (Fig. 1). Bradykinin (Fig. 2) also caused elimination of the autoregulation; this was given into the renal artery as a continuous injection in a dose of 1.5 μg/min · kg body weight.

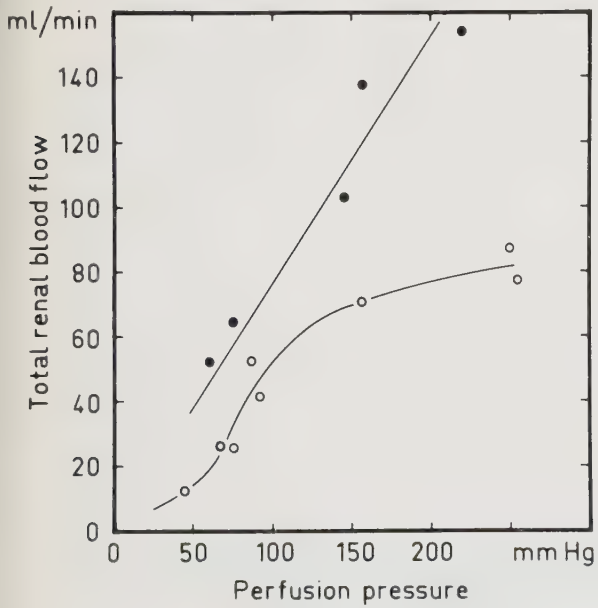
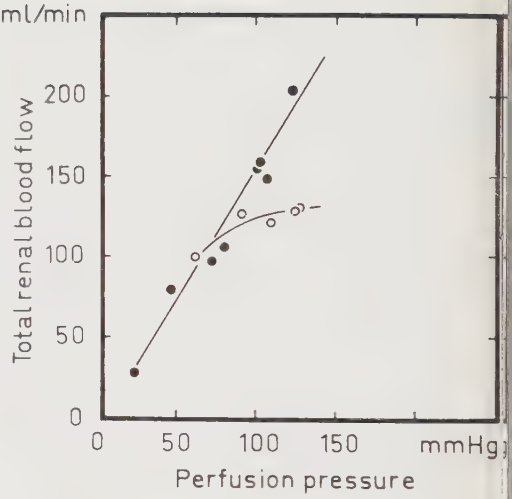


Fig. 1. The relationship between perfusion pressure and total renal blood flow, when regulated by a Sigma pump. The filled circles represent the values obtained on continuous intravenous injection of papaverine, and the unfilled circles those obtained without papaverine. The dog was anaesthetized with pentobarbital. The weight of the kidney was 31 g.

Fig. 2. The relationship between perfusion pressure and total renal blood flow, when regulated by a Sigma pump. The filled circles represent the values obtained on continuous injection of bradykinin into the renal artery, and the unfilled circles those obtained without bradykinin. The dog was anaesthetized with chloralose. The weight of the kidney was 30 g.



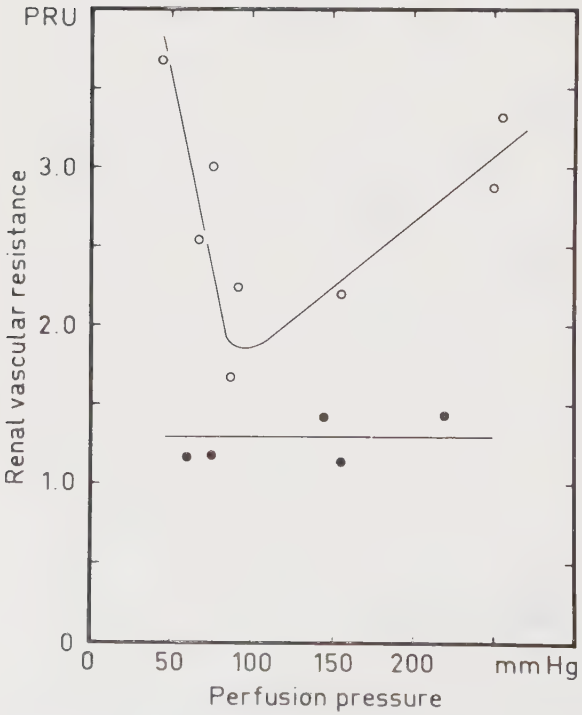


Figure 3. The figure shows the renal vascular resistance, expressed as PRU (mm Hg · min · ml^{-1}) plotted against the perfusion pressure. The values are calculated from Fig. 1. The filled circles represent values obtained with papaverine and the unfilled circles those obtained without papaverine.

When the renal vascular resistance expressed as peripheral resistance units (PRU, mm Hg · min · ml^{-1}) was plotted against perfusion pressure, Fig. 3 was obtained with values calculated from Fig. 1. With the autoregulation intact, the vascular resistance changed according to the V-shaped curve. When the autoregulation was eliminated, the vascular resistance remained constant with increasing perfusion pressure within the investigated region.

Regional autoregulation

From a chloralose-anaesthetized dog, the pressure-flow relationship shown in Fig. 4 was obtained, indicating the presence of autoregulation. The curve has a pronounced S-shape, and on increase of the perfusion pressure from 80 to 220 mm Hg the flow increased from 100 to only 120 ml/min.

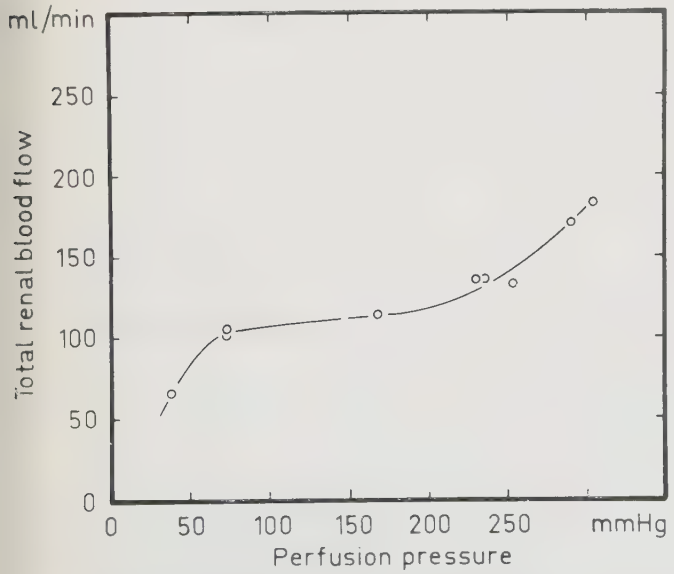


Fig. 4 A typical autoregulation curve from a chloralose-anaesthetized dog, weighing 15.5 kg. Kidney weight 35 g. The total renal blood flow remains almost constant when the perfusion pressure measured in the renal artery increases from 80 to 220 mm Hg.

When in the same dog the response from a heated thermocouple, located in the outer zone of the medulla, was plotted against the perfusion pressure the curve in Fig. 5 was obtained. The thermocouple response, $\Delta T/T_{max}$ increased only 2–3% on increase of the perfusion pressure from 80 to 220 mm Hg.

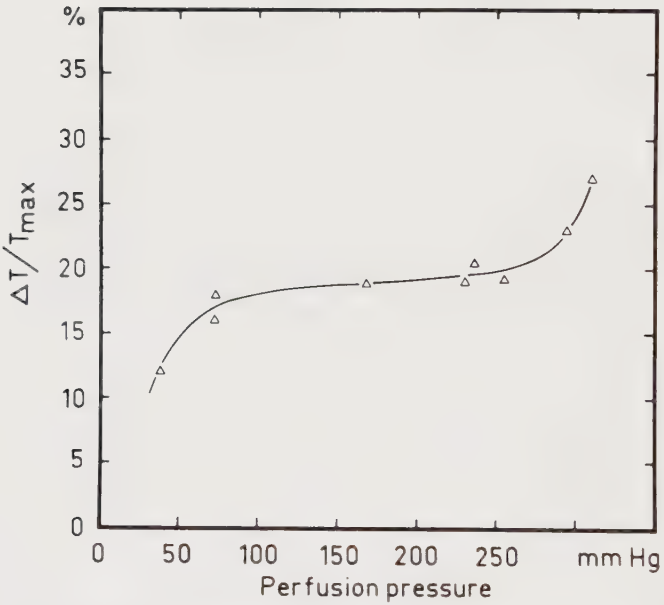


Fig. 5. The response of a thermocouple, $\Delta T/T_{max}$ located in the outer zone of the medulla, plotted against the perfusion pressure. The S-shape of the curve is pronounced and the increase in response is small when the perfusion pressure is raised from 80 to 250 mm Hg.

Plotting of the response from a heated thermocouple located in the inner zone, against the perfusion pressure, also resulted in an S-shaped curve (Fig. with only a few per cent increase in response when the perfusion pressure was raised from 80 to 220 mm Hg. When the perfusion pressure was increased further, there was a marked increase in response from the thermocouples both in the outer and inner zone of the medulla.

The diuresis increased at the high perfusion pressures of about 300 mm Hg, to approximately 3 ml/min, compared with 1.5 ml/min at perfusion pressures between 100 and 200 mm Hg.

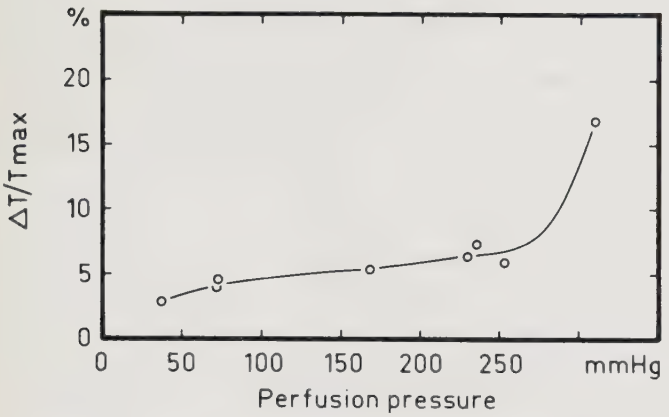


Fig. 6. The response of a thermocouple ($\Delta T/T_{max}$) located in the inner zone of the medulla, plotted against the perfusion pressure. When the perfusion is raised from 80 to 250 mm Hg, the thermocouple response increases only 2–3 per cent, but it is markedly raised when the pressure is increased further.

Clamping of the tube system

On clamping of the tube system connected to the renal artery, complete autoregulation was found down to about 80 mm Hg (Fig. 7), the total renal blood flow regaining its initial value within 3 minutes. The same pattern was found in the regional blood flow measured with heated thermocouples. When the perfusion pressure was decreased further, the regulation seemed to persist even longer in the medulla than in the cortex. Sometimes, but not regularly, a number of thermocouples in the medulla, especially those in the inner zone, even indicated increased flow at perfusion pressures between 40 and 60 mm Hg, as seen in Fig. 7.

When the constriction was released there were overshoots in the total

and regional blood flows, which did not regain the initial values until 5–6 minutes later. The urine flow which had been suppressed or stopped started again.

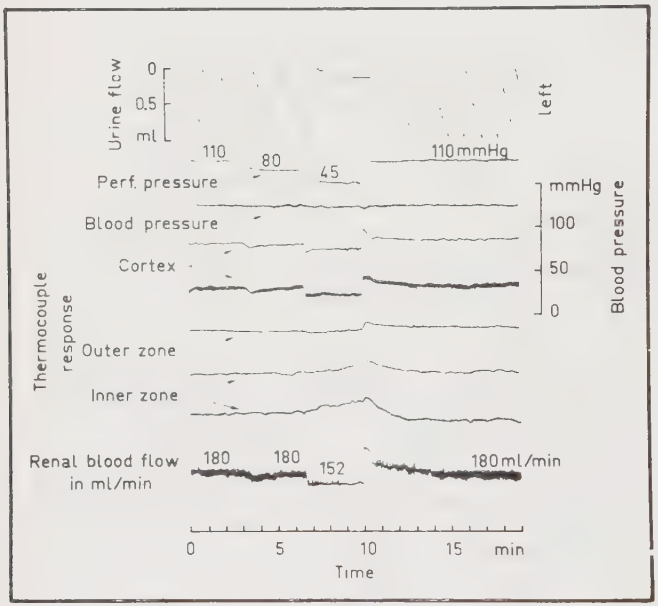


Fig. 7. The figure shows graded constriction of the extracorporeal tube system, through which the left kidney is perfused. When the perfusion pressure is decreased from 110 to 80 mm Hg the total renal blood flow, measured with a rotameter, and the regional blood flow, measured with thermocouples, are autoregulated. The urine flow is slightly diminished. Further reduction in pressure reduces the total and cortical blood flow, and the urine flow is stopped. In the medulla, however, increased blood flow is recorded. On release of the constriction an overshoot in blood flow is recorded and diuresis starts. The weight of the dog was 10.5 kg and the kidney weight 30 g.

DISCUSSION

With regard to the total flow in the kidney, the results of this study are in agreement with those of other investigators, and confirm that the kidney has a marked capacity for autoregulation of the blood flow even at considerable variations in perfusion pressure. Autoregulation of the renal blood flow was found in all animals investigated. It was possibly more marked in dogs anaesthetized with chloralose than those anaesthetized with pentobarbital, an observation in agreement with Waugh (1964).

It was found that with papaverine the autoregulation could be reduced or eliminated, which agreed with the results of Ochwadt (1956), Thurau and Kramer (1959) and Waugh and Shanks (1960), among others. The autoregulation was also eliminated by bradykinin, injected into the renal artery. This result was quite as expected, since bradykinin is a very potent relaxant for vascular musculature (Hyman and Paldino 1967), and in an earlier paper (Grängsjö and Persson 1968) it was shown that bradykinin injected into the renal artery resulted in a marked increase in the blood flow to the cortex and the medulla of the kidney.

The vascular resistance $R = P/\phi$ where P is the pressure difference (mm Hg) between the renal artery and the renal vein, and ϕ is the blood flow (ml/min) in the renal artery. When the vascular resistance for each point on the pressure-flow curve, as calculated from Fig. 4, is plottet against the perfusion pressure, Fig. 8 is obtained.

When the pressure approaches the critical closing pressure the resistance tends towards infinity. The minimum of the curve is obtained at the pressure where the autoregulation starts, and the maximum at the end of autoregulation, where the vessels are forced to dilate, as a result of increasing perfusion pressure. At the inflection point (M) the steepest part of the curve is obtained, indicating the pressure at which the kidney autoregulates maximally.

The existence of autoregulation in the cortex is well established. It is claimed that in the medulla no such mechanism exists (Thurau 1964, Moffat 1967), or only to a lesser extent (Thurau 1967). In our experiments we found an S-shaped pressure-flow relationship (actually the pressure-thermocouple response) in the outer and inner zones of the medulla, with the regional blood flow measured with heated thermocouples. It cannot be precluded, however, that the increased urine flow at the high perfusion

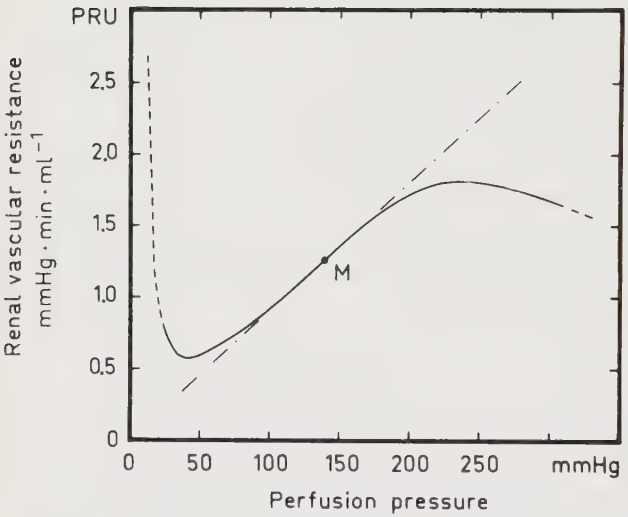


Figure 8. The relationship between the renal vascular resistance and the perfusion pressure. The minimum of the curve is at the pressure where autoregulation starts. The maximum is at the pressure, where the autoregulation ends, and the vessels are forced to dilate, as a result of increasing perfusion pressure. At the inflection point (M) the steepest part of the curve is obtained, indicating the pressure at which the kidney autoregulates maximally.

pressures could have influenced the responses from thermocouples located in the inner zone. An increase in urine flow alone can hardly explain, however, the large increase in thermocouple response, and there was probably also an increased blood flow. The existence of autoregulation in the medulla is further supported by the fact that in calibration experiments (Grängsjö 1968) it was found that the responses from thermocouples in the medulla increased with increasing flows in the renal artery when the autoregulation was eliminated with papaverine.

These findings, the S-shaped pressure-thermocouple response relationship and the difference between calibration curves with the presence or absence of papaverine, we have interpreted as the existence of autoregulation in the renal medulla. With regard to the outer zone, this opinion is supported by Aukland (1967) and Wolgast (1968). Thurau et al. (1960) considered, however, that there was no autoregulation in the renal medulla. These differences

of opinion can possibly be explained by the different experimental conditions, such as the type and depth of anaesthesia and the degree of traumatic effect of the different methods of measurement on the normal flow conditions in the medulla. It is also conceivable that the electrolyte status of the experimental animals may have varied, which is of importance since the plasma osmolality and the sodium load can influence the autoregulation (Guyton et al. 1966, Thurau 1967), via the juxtaglomerular apparatus and the renin-angiotensin system.

On graded decrease of the perfusion pressure by clamping of the tube system, complete autoregulation was found in the cortex and medulla down to 80 mm Hg. On some occasions, but not regularly, the medulla appeared to autoregulate to even lower perfusion pressures than the cortex. Somewhat confusing was the increased response which was sometimes found in the medulla, and especially in the inner zone, at perfusion pressures between 40 and 60 mm Hg. The diuresis had then stopped and thus had no influence on the thermocouple response. The recorded increase may well therefore have been true and could have been due to an increased medullary blood flow. These findings are in concordance with the theory of redistribution of intrarenal blood flow on hypotension, proposed by Trueta et al. (1947), and results presented recently by Carriere et al. (1966), but require further investigation. It is not inconceivable, however, that under certain circumstances the vessels in the cortex and medulla can react differently, in view of the essential differences between them as regards the innervation of their smooth muscle (Coupland 1962, Nilsson 1965, Moffat 1967).

On release of the constriction overshoots were found in the total and regional blood flows (cortex and medulla). After 5–6 minutes these had returned to the initial values. This increase in total renal blood flow on release of a constriction has been demonstrated previously (Waugh and Shanks 1960, Schmid and Spencer 1962, Thurau and Henne, cited Thurau 1964), and is said to reflect the decreased tone of the vascular smooth muscle which develops during the obstruction period. During the minutes immediately following the release the vascular tone will increase and the blood flow will be regulated. The mechanism of this overshoot is postulated to be wholly myogenic in origin (Grupp and Heimpel 1958, Schmid and Spencer 1962). According to other authors, however, (e.g. Waugh 1964), local metabolic components may possibly have a role in this regulation of blood flow.

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